Heart Failure

Multimarker Approach for the Prediction of Heart Failure Incidence in the Community

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Background—Several biological pathways are activated in ventricular remodeling and in overt heart failure (HF). There are no data, however, on the incremental utility of a parsimonious set of biomarkers (reflecting pathways implicated in HF) for predicting HF risk in the community.

Methods and Results—We related a multimarker panel to the incidence of a first HF event in 2754 Framingham Heart Study participants (mean age, 58 years; 54% women) who were free of HF and underwent routine assays for 6 biomarkers (C-reactive protein, plasminogen activator inhibitor-1, homocysteine, aldosterone-to-renin ratio, B-type natriuretic peptide, and urinary albumin-to-creatinine ratio). We estimated model c statistic, calibration, and net reclassification improvement to assess the incremental predictive usefulness of biomarkers. We also related biomarkers to the incidence of nonischemic HF in participants without prevalent coronary heart disease. On follow-up (mean, 9.4 years), 95 first HF events occurred (54 in men). In multivariable-adjusted models, the biomarker panel was significantly related to HF risk (P=0.00005). On backward elimination, B-type natriuretic peptide and urinary albumin-to-creatinine ratio emerged as key biomarkers predicting HF risk; hazards ratios per 1-SD increment in log marker were 1.52 (95% confidence interval, 1.24 to 1.87) and 1.35 (95% confidence interval, 1.11 to 1.66), respectively. B-type natriuretic peptide and urinary albumin-to-creatinine ratio significantly improved the model c statistic from 0.84 (95% confidence interval, 0.80 to 0.88) in standard models to 0.86 (95% confidence interval, 0.83 to 0.90), enhanced risk reclassification (net reclassification improvement=0.13; P=0.002), and were independently associated with nonischemic HF risk.

Conclusion—Using a multimarker strategy, we identified B-type natriuretic peptide and urinary albumin-to-creatinine ratio as key risk factors for new-onset HF with incremental predictive utility over standard risk factors. (Circulation. 2010;122:1700-1706.)

Key Words: biomarkers ■ epidemiology ■ heart failure ■ natriuretic peptides ■ risk ■ prediction

Heart failure (HF) is associated with high morbidity and mortality, making its prevention a public health priority. Identification of people who are at higher risk of developing HF is critical for targeting prevention strategies. Investigators from the Framingham Heart Study previously described an HF “risk profile” based on clinical, ECG, and x-ray features, but these clinical factors do not fully explain HF risk. Recently, numerous investigations have highlighted that several biological pathways are activated during left ventricular (LV) remodeling and HF evolution. Several reports focused on individual circulating and urinary biomarkers representing some of these key pathways, but few have assessed the incremental predictive utility of multiple biomarkers considered together.

Clinical Perspective on p 1706

We recently applied a multimarker strategy to identify key biomarkers associated with indexes of LV remodeling and vascular stiffness. In the present investigation, we extend the multimarker strategy to overt HF by relating the panel of
biomarkers to the incidence of a first HF event in a large community-based sample. The biomarker panel included aldosterone-to-renin ratio (ARR; renin-angiotensin-aldosterone axis), C-reactive protein (CRP; inflammation), plasminogen activator inhibitor-1 (PAI-I; fibrinolysis), B-type natriuretic peptide (BNP; natriuretic peptide system), homocysteine (oxidative stress), and the urine albumin-to-creatinine ratio (UACR; endothelial function). We hypothesized that 1 or more of these circulating and urinary biomarkers will be associated with HF risk and will incrementally predict HF incidence beyond established risk factors.

Methods

Study Sample
Details of the Framingham Offspring Study have been published previously. In brief, 5124 individuals who were children (or spouses of children) of the original Framingham cohort participants were enrolled in 1971 in the Framingham Offspring Study, and these individuals have been examined approximately every 4 years. For the present investigation, we included attendees at the sixth examination cycle (1995 to 1998; n=3532), referred to as the baseline examination for our analysis. Of these, we excluded 737 participants because of unavailable biomarker information, 10 participants for missing covariate information, and another 31 participants who had prevalent HF. Thus, 2754 participants (54% women) remained eligible for this investigation. All participants provided written informed consent, and the Institutional Review Board of the Boston University Medical Center approved the study protocol.

Measurement of Biomarkers
Biosamples were obtained on the morning of the baseline examination (when covariate information was also collected and follow-up started) after an overnight fast, usually between 8 and 9 AM, and frozen at −80°C without any freeze-thaw cycles until assays were performed. Plasma PAI-1 was determined with an ELISA test for PAI-1 antigen (TintElize PAI-1, Biopool, Ventura, Calif). CRP was measured with the Dade-Behring BN100 nephelometer. Serum aldosterone was measured with a radioimmunoassay (Quest Diagnostics, Cambridge, Mass), and plasma renin concentrations were measured by an immunochromoluminometric assay (Nichols, San Juan Capistrano, Calif). Plasma BNP was measured with a high-performance liquid chromatography assay (Shionogi, Osaka, Japan). We used high-performance liquid chromatography with fluorometric detection to measure total plasma homocysteine. We assessed UACR on a morning urine specimen using immunoturbidimetry (Tina-Quant Albumin Assay, Roche Diagnostics, Indianapolis, Ind) to measure urine albumin and the modified Jaffe method to measure urinary creatinine.

The following were the average interassay coefficients of variation for the biomarker measurements: PAI-1, 7.7%; CRP, 2.2%; renin, 2.0% (high concentrations) to 10% (low concentrations); aldosterone, 4.6% (high concentrations) to 9.8% (low concentrations); BNP, 12.2%; homocysteine, 9%; urine albumin, 7.2%; and urine creatinine, 2.3%.

HF Assessment
Follow-up for the present investigation extended from the baseline examination through December 2007. An end-points adjudication committee consisting of 3 physicians evaluated all suspected cardiovascular disease events (including HF) by reviewing Heart Study clinic charts and hospitalization and physician office records and ascertained the incidence of events according to criteria described previously. We used Framingham HF criteria to adjudicate HF incidence. In secondary analyses, we referred to participants who developed HF without an interim myocardial infarction (MI) or unstable angina (also known as coronary insufficiency) as the “nonischemic” HF group for simplicity.

Statistical Analyses
Biomarker values were natural logarithmically transformed (to account for skewed distributions) and standardized within sex to account for sex-related differences in biomarker distributions. We modeled aldosterone and renin together as a ratio, the ARR, because in our cohort such combined modeling has been most informative. We calculated age- and sex-adjusted Spearman coefficients to evaluate correlations between biomarkers.

To evaluate the predictive utility of biomarkers (with regard to HF risk), we performed the following analyses. First, after confirming that the assumption of proportionality of hazards was met, we fitted Cox models that incorporated age, sex, body mass index, systolic blood pressure, hypertension treatment, diabetes mellitus, ratio of total cholesterol to high-density lipoprotein cholesterol, smoking, prevalent MI, and valvular heart disease. Next, we tested whether the set of 6 biomarkers (denoted m1 through m6 in the equation) was associated with HF risk using a 6-df likelihood ratio test of the null hypothesis H0: βm1=βm2=...=βm6=0. The likelihood ratio test statistic in the latter. Second, we calculated the c statistic for the model with several biomarkers in any individual, we constructed a weighted biomarker score (including only biomarkers that emerged in backward elimination): Biomarker score (β coefficient estimate of biomarker1 sex-standardized log-biomarker1 concentration)+(β coefficient estimate of biomarker2 sex-standardized log-biomarker2 concentration), etc.

We plotted the cumulative incidence of HF (accounting for competing risk of mortality) according to tertiles of the biomarker score, ascertained HF event proportions in each tertile, and evaluated whether participants in the second and third tertiles had higher HF risk compared with those in the first tertile in the multivariable-adjusted models described above. We also repeated these analyses in a subsample with Framingham Risk Score–predicted baseline 10-year coronary heart disease (CHD) risk ≥10%. In addition, we used the top tertile of the biomarker score as a threshold to calculate the sensitivity, specificity, positive predictive value, negative predictive value, likelihood ratio of positive test, and likelihood ratio of negative test to evaluate the performance characteristics of the score for possible HF risk screening.

To assess the utility of biomarkers associated with HF, we evaluated several metrics. First, we assessed model calibration (ie, concordance of observed risk and that predicted by the model with biomarkers) by calculating the Hosmer-Lemeshow Χ² statistic for Cox models. Second, we calculated the c statistic for the model with clinical covariates alone and compared it with that for the model with clinical covariates and biomarkers to estimate the increase in the c statistic in the latter. Third, we classified participants into 3 categories of absolute 12-year HF risk (the maximum follow-up duration): low risk (<2%), intermediate risk (2% to 8%), and high risk (≥8%). We evaluated whether inclusion of biomarkers improved risk classification of participants by calculating the net reclassification improvement (NRI). The NRI is used to assess how well a new marker “reclassifies” people from 1 risk category to another (higher or lower). It is calculated as a sum of 2 separate components (1 for individuals with events and 1 for individuals without events) as follows: NRI=[(events reclassified higher−events reclassified lower)/events]+[(nones reclassified lower−nones reclassified higher)/nones].
Table 1. Baseline Clinical and Biochemical Characteristics of Study Participants

<table>
<thead>
<tr>
<th>Variable</th>
<th>Men (n=1278)</th>
<th>Women (n=1476)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Clinical characteristics</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age, y</td>
<td>59 (10)</td>
<td>58 (10)</td>
</tr>
<tr>
<td>Body mass index, kg/m²</td>
<td>28.7 (4.4)</td>
<td>27.4 (5.8)</td>
</tr>
<tr>
<td>Systolic blood pressure, mm Hg</td>
<td>130 (17)</td>
<td>127 (20)</td>
</tr>
<tr>
<td>Ratio of total to high-density lipoprotein cholesterol</td>
<td>4.9 (2.0)</td>
<td>3.9 (1.4)</td>
</tr>
<tr>
<td>Hypertension treatment, %</td>
<td>30</td>
<td>25</td>
</tr>
<tr>
<td>Current smoking, %</td>
<td>14</td>
<td>16</td>
</tr>
<tr>
<td>Diabetes mellitus, %</td>
<td>12</td>
<td>9</td>
</tr>
<tr>
<td>Valvular heart disease, %</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>Prevalent MI, %</td>
<td>7</td>
<td>1</td>
</tr>
<tr>
<td><strong>Biochemical characteristics</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CRP, mg/L</td>
<td>1.81 (0.90, 3.91)</td>
<td>2.40 (0.99, 5.63)</td>
</tr>
<tr>
<td>PAI-1, ng/mL</td>
<td>25.6 (17.1, 36.0)</td>
<td>20.2 (12.1, 31.8)</td>
</tr>
<tr>
<td>Homocysteine, mmol/L</td>
<td>9.81 (8.26, 11.92)</td>
<td>8.30 (6.97, 10.13)</td>
</tr>
<tr>
<td>ATR</td>
<td>0.65 (0.38, 1.14)</td>
<td>1.00 (0.55, 1.67)</td>
</tr>
<tr>
<td>BNP, pg/mL</td>
<td>6.10 (4.00, 15.9)</td>
<td>9.70 (4.00, 19.65)</td>
</tr>
<tr>
<td>Urine albumin/creatinine ratio, mg/g</td>
<td>4.88 (2.15, 10.93)</td>
<td>8.55 (3.57, 17.24)</td>
</tr>
</tbody>
</table>

Values are mean (SD) for clinical characteristics and median (first quartile, third quartile) for biochemical parameters.

Results

Baseline clinical and biochemical characteristics of our sample are displayed in Table 1. Of the 2754 participants in our sample, 118 had a history of MI or unstable angina at baseline. Approximately half the participants in our sample had a baseline Framingham Risk Score–predicted 10-year CHD risk ≥10%, and a fifth had predicted 10-year CHD risk ≥20%. Of note, women made up ≈70% of the group with 10-year CHD risk <10%. Approximately 80% of the HF events were concentrated in the subgroup of participants with Framingham Risk Score–predicted 10-year CHD risk ≥10%. Age- and sex-adjusted correlations among the biomarkers are presented in Table II in the online-only Data Supplement. The strongest positive and inverse correlations were between CRP and PAI-1 and between PAI-1 and BNP, respectively (Table II in the online-only Data Supplement).

Over a mean follow-up of 9.4 years (maximum, 12.8 years), 95 participants (41 women) developed HF. The panel of biomarkers was significantly related to HF risk (P=0.00005). On backward elimination, BNP and UACR emerged as significant correlates; each 1-SD increase in log BNP and log UACR was associated with a 15% and 35% higher risk of developing HF, respectively (Table 2). When BNP and UACR were modeled together as a biomarker score, unadjusted HF proportions increased 10-fold across tertiles. Cumulative HF incidence curves by tertile of biomarker score are presented in the Figure. Participants in the second and third tertiles of biomarker score carried multivariable-adjusted HF hazards that were 3-fold and 4-fold higher, respectively, than those with a biomarker score in the first tertile (Table 3). Results were similar when we repeated these analyses in a subsample with Framingham Risk Score–predicted baseline 10-year CHD risk ≥10% (Table III in the
### Online-only Data Supplement

The sensitivity, specificity, positive predictive value, negative predictive value, likelihood ratio of positive test, and likelihood ratio of negative test of the top tertile of biomarker score were 68%, 68%, 7%, 98%, 2.13, and 0.46, respectively. When these metrics were recalculated using a sample of individuals with at least 6 years of follow-up, the estimates were very similar.

The addition of biomarkers to the model with clinical characteristics improved the model c statistic from 0.84 (95% confidence interval [CI], 0.80 to 0.88) to 0.86 (95% CI, 0.83 to 0.90; $P=0.007$ for improvement), and the model with biomarkers had excellent calibration (Hosmer-Lemeshow $\chi^2=9.45; P=0.40$). The contributions of BNP and UACR to the improvement in c statistic were of equal magnitude (0.01 each); c statistics for models with clinical covariates alone, with covariates and BNP, and with covariates and BNP and UACR were 0.84, 0.85, and 0.86, respectively. When participants who developed HF and those who did not were classified separately into risk categories based on clinical characteristics alone, the addition of biomarkers reclassified 13% of participants in the appropriate direction; ie, those without HF on follow-up in the intermediate-risk group were reclassified “downward,” and those with HF on follow-up in the intermediate-risk group were reclassified “upward” (NRI=0.13; $P=0.002$; Table 4).

In secondary analyses, relations of BNP and UACR to HF risk remained robust on adjustment for interim MI (Table 2). Results of the jackknife cross-validation showed that the basic model not including the 2 biomarkers produced a jackknife c statistic of 0.838 (95% CI, 0.797 to 0.879), whereas addition of the 2 markers to the basic model produced a jackknife c statistic of 0.862 (95% CI, 0.825 to 0.899), yielding a jackknife overoptimism estimate of 0.024 (95% CI, 0.007 to 0.041; $P=0.007$), therefore suggesting only a modest degree of overoptimism. In the subsample of participants without baseline MI or unstable angina (n=2636), 57 developed nonischemic HF on follow-up. In these analyses, each 1-SD increment in log BNP and log UACR was associated with a 78% ($P<0.001$) and 39% ($P=0.02$) higher HF risk, respectively (Table 2). When analyses evaluating the relations of biomarker score to HF risk were adjusted for all incident and prevalent ischemic events (MI, unstable angina, and angina pectoris), the results were unchanged (Table IV in the online-only Data Supplement). Additional adjustment of the multivariable model with biomarker score for incident and prevalent unrecognized MI did not alter the results (Table V in the online-only Data Supplement). In addition, results did not differ when our analyses were repeated in a subgroup of participants with normal LV systolic function (data not shown).

Relations of BNP and UACR to HF risk were not altered by additional adjustment for estimated glomerular filtration rate and were similar in participants with estimated glomerular filtration rate $\geq 60$ mL min$^{-1}$1.73 m$^{-2}$ (data not shown). Relations of biomarker score tertiles to HF risk were unchanged when clinical risk factors were modeled as time-dependent covariates (Table VI in the online-only Data Supplement). Finally, relations of BNP and UACR to HF risk were not modified by age, sex, or body mass index (None of the interaction terms was statistically significant).

### Table 3. Relations of Biomarker Score to HF Risk

<table>
<thead>
<tr>
<th>Events-at Risk, n (%)</th>
<th>Tertile 1</th>
<th>Tertile 2</th>
<th>Tertile 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Event proportions by tertile of biomarker score based on BNP and UACR.</td>
<td>6/918 (0.7)</td>
<td>24/918 (2.6)</td>
<td>65/918 (7.1)</td>
</tr>
<tr>
<td>$P=0.02$</td>
<td>2.9 (1.7–7.0)</td>
<td>4.2 (1.8–10.2)</td>
<td></td>
</tr>
</tbody>
</table>

**Event proportions by tertile of biomarker score based on BNP and UACR.**

### Discussion

#### Principal Findings

In our large community-based sample, we identified BNP and UACR (from a multimarker panel) as key predictors of HF risk, emphasizing the importance of natriuretic peptide system activation and endothelial dysfunction as markers of disease progression. BNP and UACR were also indepen-

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**Table 2. Classification of Participants Into HF Risk Groups Based on Multivariable Models With and Without Biomarkers**

<table>
<thead>
<tr>
<th>Risk</th>
<th>Total</th>
<th>Risk reclassification in participants with HF, n (row %)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;2%</td>
<td>1644</td>
<td>2%–8% 0.001</td>
</tr>
<tr>
<td>2%–8%</td>
<td>765</td>
<td>&gt;8% 0.02</td>
</tr>
<tr>
<td>&gt;8%</td>
<td>250</td>
<td>Total 2659</td>
</tr>
</tbody>
</table>

NRI=[[(7+8)−(3+3)95+[(176+74)−(115+49)2659=0.13; $P=0.009$. $\text{HF risk categories based on clinical covariates plus biomarker score.}$

$\text{HF risk categories based on clinical covariates alone.}$
dentily associated with nonischemic HF, suggesting that relations of biomarkers to HF risk are not mediated solely by interim occurrence of ischemic events, and were significantly associated with HF risk in the subgroup with normal LV systolic function, suggesting that our findings were not driven by individuals with LV systolic dysfunction in our sample. In addition, participants who were excluded because of intercurrent ischemic events as having “ischemic HF” may still have nonischemic HF (ie, the intervening event may not be causally related to HF occurrence). These 2 biomarkers improved HF risk prediction very modestly, as evidenced by improvements in model discrimination and risk reclassification. The biomarker score may have potential utility as a screening tool, a premise that would require additional studies; the high negative predictive value may be important to note in this context. We also demonstrate the robustness of the biomarkers in predicting HF risk in participants with high baseline CHD risk; however, we should be cautious in generalizing these findings to people with low baseline CHD risk (10-year risk <10%), and biomarker utility in the latter group needs further study.

Several previous investigations reported the relations of biomarkers from various biological domains to HF risk. However, our investigation is novel in several respects. Whereas earlier studies evaluated biomarkers individually, we used a multimarker strategy, which permitted a comparison of several biomarkers while limiting multiple statistical testing. We also assessed the potential incremental utility of biomarkers (and biomarker scores) for predicting HF risk (above and beyond standard clinical risk factors). An additional strength of our report is the demonstration that both BNP and UACR are associated with the risk of developing nonischemic HF in a large subgroup without history of MI or unstable angina, thereby avoiding potential confounding by preexisting and interim ischemic events, which can activate several of the pathways represented by the biomarkers we investigated.

Although the exact mechanisms underlying the predictive value of BNP and UACR cannot be conclusively determined from epidemiological data alone, the results from our investigation, if confirmed, can potentially be used in clinical settings to evaluate HF risk and to identify specific high-risk individuals. Analogous to the use of multivariable risk profiles in the assessment of CHD risk in people with dyslipidemia, with attendant determination of treatment targets, biomarker risk scores can be used to identify those who are at high risk for HF and may potentially benefit from treatment of currently established HF risk factors (hypertension, diabetes, obesity, etc) to targets that are lower than conventionally recommended to reduce HF incidence. However, this approach (using biomarker scores for risk stratification) has yet to be tested and need validation before use in clinical settings. Indeed, it may be argued that all patients at risk of developing HF warrant aggressive management of their risk factor burden.

Mechanisms Underlying the Relations of Biomarkers to HF Risk

BNP is a hormone with natriuretic, diuretic, and vasodilatory properties that is released in response to increased LV filling pressures and/or greater LV wall stress. BNP has been used in the diagnosis of clinical HF but is also elevated in people with asymptomatic LV dysfunction. Thus, one explanation for the relations of BNP to HF risk is that participants with elevated BNP concentrations are those with subclinical LV remodeling, systolic dysfunction, or diastolic dysfunction and therefore develop HF at higher rates. Another possibility is that relations of BNP to HF risk are mediated by its relations to incident ischemic cardiovascular events. BNP may be released in the setting of ischemic events other than MI or with subclinical ischemia and may therefore predict HF secondary to clinical or subclinical ischemia. In our investigation, a variety of adjustments for ischemic events (including adjustment for unrecognized MI) and exclusion of participants with MI and unstable angina did not alter the results. However, it is possible that BNP released in response to subclinical ischemia may be an explanation for the association between this biomarker and HF risk. Overall, it is likely that BNP is a marker for each of the mechanisms noted above.

Similarly, UACR is a risk marker for endothelial dysfunction, target organ damage, ischemic events, and an atherogenic risk profile, which may explain why it is related to HF risk. As with BNP, it is likely that UACR is a cumulative measure for all these mechanisms for HF risk.

The other biomarkers in our panel (CRP, PAI-1, ARR, and homocysteine) have previously been implicated in ventricular remodeling, alterations in LV function, and HF incidence. However, in our analysis, we did not observe an independent association between these biomarkers and HF risk. It is conceivable that the relations of these biomarkers to HF risk may be mediated through their relations to clinical risk factors that we adjusted for in our model. Indeed, previous reports have described the associations between CRP and hypertension and diabetes mellitus and between PAI-1 and hypertension and metabolic syndrome. Prior investigations also have noted the relations of ARR to hypertension and both ARR and homocysteine to vascular stiffness. Thus, our results do not imply that the pathways represented by these biomarkers do not contribute to HF risk.

Strengths and Limitations

Our study is strengthened by a large sample size, standardized measurements of biomarkers and clinical variables, a rigorous definition of HF events, and a conservative analysis strategy to minimize multiple testing. However, several limitations should be acknowledged. First, we tested only a small set of biomarkers that were available at a routine examination and that have previously been implicated in LV remodeling and/or HF risk; markers of other biological pathways (or other biomarkers of domains we evaluated) that were not tested may be important in influencing HF risk. Second, differences in the analytic precision of the assays for these biomarkers may have influenced the results of analysis. Third, we lack information on quantitative LV ejection fraction, measures of diastolic function, and measures of endothelial or vascular function at the baseline examination; therefore, we could not adjust for these measures in our multivariable models. Fourth, reclassification metrics and
performance characteristics of the biomarker score are susceptible to misclassification of participants (cases versus noncases) in whom follow-up information is not complete. Finally, our sample comprised middle-aged white individuals of European ancestry, and our results may not be generalizable to other age groups or ethnicities.

Conclusions
Refining HF prediction is a fundamental step for preventing the condition. In our prospective investigation of a large community-based sample of middle-aged whites, we identified BNP and UACR as key biomarkers associated with HF risk. However, the incremental usefulness of these biomarkers over standard clinical factors (as assessed by c statistics and NRI) is very modest. Additional investigations are therefore warranted both to replicate our findings and to assess their applicability to routine clinical settings.

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Disclosures
None.

References
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SUPPLEMENTAL MATERIAL: A Multimarker Approach for Prediction of Heart Failure Incidence in the Community.

I. Definitions of covariates

II. Supplementary Table 1. Framingham heart failure criteria

III. Statistical methods for secondary analyses

IV. Supplementary Table 2. Correlations between biomarkers

V. Supplementary Table 3: Analyses confined to high-risk participants

VI. Supplementary Table 4: Analyses adjusting for all clinical ischemic events

VII. Supplementary Table 5: Analyses adjusting for unrecognized MI

VIII. Supplementary Table 6: Analyses modeling clinical risk factors as time-dependent covariates
I. Definitions of covariates

Covariates used for multivariable analyses were all defined at the baseline examination. We calculated body mass index (BMI) as the weight in kilograms divided by the square of height in meters. A physician measured blood pressure twice on the left arm of the seated participants using a mercury-column sphygmomanometer and a cuff of appropriate size; the average of two such readings was considered the examination blood pressure. Fasting lipids and glucose were measured using standardized assays. We defined diabetes as a fasting plasma glucose $\geq 126$ mg/dl or receiving hypoglycemic therapy. We defined valve disease as $\geq$moderate stenosis or regurgitation of the mitral or the aortic valves on Doppler color flow imaging at the echocardiographic examination performed at the baseline examination. Interim MI or unstable angina was defined as occurrence of these events between baseline examination and time of HF occurrence.
## II. Supplementary Table 1. Framingham Criteria for HF Diagnosis

<table>
<thead>
<tr>
<th>Major Criteria</th>
<th>Minor Criteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Paroxysmal nocturnal dyspnea or orthopnea</td>
<td>Dyspnea on ordinary exertion</td>
</tr>
<tr>
<td>Distended neck veins</td>
<td>Night cough</td>
</tr>
<tr>
<td>Hepatojugular reflux</td>
<td>Heart rate &gt; 120/minute</td>
</tr>
<tr>
<td>Rales</td>
<td>Hepatomegaly</td>
</tr>
<tr>
<td>S3 gallop</td>
<td>Ankle edema</td>
</tr>
<tr>
<td>Enlarged heart by X-ray</td>
<td>Decrease in vital capacity by 1/3rd</td>
</tr>
<tr>
<td>Acute pulmonary edema on chest X-ray</td>
<td>Pleural effusion by X-ray</td>
</tr>
<tr>
<td>Treatment induced weight loss &gt;10lbs/5 days</td>
<td>Pulmonary vascular engorgement by X-ray</td>
</tr>
<tr>
<td>Increased venous pressure &gt; 16 cm water</td>
<td></td>
</tr>
<tr>
<td>Pulmonary edema, visceral congestion or cardiomegaly on autopsy</td>
<td></td>
</tr>
</tbody>
</table>

HF is diagnosed when 1 major and 2 minor criteria or 2 major criteria are present without a competing explanation for findings.
III. Statistical Methods for Secondary Analyses

We performed several secondary analyses on the biomarkers identified in our primary analyses as predictors of HF risk. Several biomarkers in our panel have been previously related to CVD risk,¹ and they may, therefore, influence HF risk via ischemic events during follow-up. We examined this possibility in four different ways. First, we included interim MI as a time-dependent covariate in our primary models relating the biomarkers to HF risk. Second, we related biomarkers to risk of developing non-ischemic HF in a subgroup of participants without history of major coronary disease (MI or unstable angina) at baseline; for these analyses, participants who developed an MI or unstable angina on follow-up were censored as non-events at the time of the ischemic event. Third, we repeated the analyses evaluating relations of the biomarker score to HF risk adjusting for all ischemic events (MI, unstable angina and angina pectoris), both incident and prevalent. Fourth, we also repeated the multivariable model with biomarker score additionally adjusting for incident and prevalent unrecognized MI.

The relations of biomarkers to HF risk may be mediated by their relations to underlying renal function, so we (a) additionally adjusted our primary model for estimated glomerular filtration rate (eGFR) as calculated by the “Modification of Diet in Renal Disease” formula,² and (b) repeated our primary model in a sub-sample with eGFR ≥60 ml/min/1.73 m². As values for covariates (such as BMI) and proportions of participants with co-existing heart disease (such as valve disease) may change over time, we repeated the multivariable model with biomarker score treating all clinical risk factors as time-dependent covariates. Lastly, since BNP³,⁴ and UACR⁵,⁶ values vary with age, sex, and BMI we tested if relations of biomarkers to HF risk varied according to these covariates by incorporating first-order interaction terms (age dichotomized at
median; sex, men versus women; BMI dichotomized at <30 versus ≥30 kg/m²) in the multivariable models.
IV. Supplementary Table 2. Correlations among Biomarkers

<table>
<thead>
<tr>
<th></th>
<th>CRP</th>
<th>PAI-1</th>
<th>Homocysteine</th>
<th>ARR</th>
<th>BNP</th>
<th>UACR</th>
</tr>
</thead>
<tbody>
<tr>
<td>CRP</td>
<td>1.00</td>
<td></td>
<td>0.03</td>
<td>0.02</td>
<td>-0.05</td>
<td>0.02</td>
</tr>
<tr>
<td>PAI-1</td>
<td>1.00</td>
<td>0.06</td>
<td></td>
<td>0.00</td>
<td>-0.14</td>
<td>0.00</td>
</tr>
<tr>
<td>Homocysteine</td>
<td>1.00</td>
<td></td>
<td>-0.06</td>
<td>0.06</td>
<td></td>
<td>0.04</td>
</tr>
<tr>
<td>ARR</td>
<td></td>
<td></td>
<td></td>
<td>1.00</td>
<td>0.09</td>
<td></td>
</tr>
<tr>
<td>BNP</td>
<td></td>
<td></td>
<td></td>
<td>1.00</td>
<td></td>
<td>0.06</td>
</tr>
<tr>
<td>UACR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1.00</td>
</tr>
</tbody>
</table>

Cells display age- and sex-adjusted Spearman partial correlation coefficients for natural log-transformed and sex-standardized mean biomarker values, calculated on a sample of 2754 individuals. Values in bold indicate correlations that are statistically significant at a p-value threshold of 0.05.

CRP = C-reactive protein; PAI-1 = plasminogen activator inhibitor-1; ARR = aldosterone-to-renin ratio; BNP = B-type natriuretic peptide; UACR = urinary albumin-to-creatinine ratio.
V. Supplementary Table 3. Relations of Biomarker Score to HF Risk in Participants with Predicted 10-year CHD Risk ≥ 10%

<table>
<thead>
<tr>
<th></th>
<th>Tertile 1</th>
<th>Tertile 2</th>
<th>Tertile 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of events/no. at risk (%)*</td>
<td>5/367 (1.4)</td>
<td>18/449 (4.1)</td>
<td>56/540 (10.4)</td>
</tr>
<tr>
<td>HF risk†</td>
<td>Referent</td>
<td>2.36 (0.87 – 6.38)</td>
<td>3.91 (1.50 – 10.18)</td>
</tr>
</tbody>
</table>

Results based on sub-sample of participants with predicted 10-year CHD risk ≥ 10% as assessed by Framingham Risk Score.

* Event proportions by tertile of biomarker score based on BNP and UACR

† Multivariable-adjusted HF hazards (confidence intervals) compared to referent group. Model adjusted for age, sex, body mass index, systolic blood pressure, hypertension treatment, diabetes, current smoking, total/HDL cholesterol ratio, valvular heart disease and prevalent myocardial infarction.
VI. Supplementary Table 4. Relations of Biomarker Score to HF Risk: Adjustment for all Clinical Ischemic Events

<table>
<thead>
<tr>
<th></th>
<th>Tertile 1</th>
<th>Tertile 2</th>
<th>Tertile 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of events/no. at risk (%)*</td>
<td>6/918 (0.7)</td>
<td>24/918 (2.6)</td>
<td>65/918 (7.1)</td>
</tr>
<tr>
<td>HF risk†</td>
<td>Referent</td>
<td>2.69 (1.09 – 6.61)</td>
<td>4.39 (1.84 – 10.48)</td>
</tr>
</tbody>
</table>

* Event proportions by tertile of biomarker score based on BNP and UACR

† Multivariable-adjusted HF hazards (confidence intervals) compared to referent group. Model adjusted for age, sex, body mass index, systolic blood pressure, hypertension treatment, diabetes, current smoking, total/HDL cholesterol ratio, valvular heart disease and both prevalent and incident ischemic events (MI, unstable angina and angina pectoris).
VII. Supplementary Table 5. Relations of Biomarker Score to HF Risk: Additional Adjustment for Unrecognized MI

<table>
<thead>
<tr>
<th>Tertile 1</th>
<th>Tertile 2</th>
<th>Tertile 3</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>No. of events/no. at risk (%)</strong></td>
<td>6/918 (0.7)</td>
<td>24/918 (2.6)</td>
</tr>
<tr>
<td><strong>HF risk†</strong></td>
<td>Referent</td>
<td>2.69 (1.10 – 6.62)</td>
</tr>
</tbody>
</table>

* Event proportions by tertile of biomarker score based on BNP and UACR

† Multivariable-adjusted HF hazards (confidence intervals) compared to referent group. Model adjusted for age, sex, body mass index, systolic blood pressure, hypertension treatment, diabetes, current smoking, total/HDL cholesterol ratio, valvular heart disease, prevalent recognized MI and prevalent and incident unrecognized MI.
VIII. Supplementary Table 6. Relations of Biomarker Score to HF Risk: Clinical Risk Factors Modeled as Time-Dependent Covariates

<table>
<thead>
<tr>
<th></th>
<th>Tertile 1</th>
<th>Tertile 2</th>
<th>Tertile 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of events/no. at risk (%)*</td>
<td>6/918 (0.7)</td>
<td>24/918 (2.6)</td>
<td>65/918 (7.1)</td>
</tr>
<tr>
<td>HF risk†</td>
<td>Referent</td>
<td>2.73 (1.11 – 6.71)</td>
<td>4.33 (1.81 – 10.34)</td>
</tr>
</tbody>
</table>

* Event proportions by tertile of biomarker score based on BNP and UACR

† Multivariable-adjusted HF hazards (confidence intervals) compared to referent group. Model adjusted for age, sex, body mass index, systolic blood pressure, hypertension treatment, diabetes, current smoking, total/HDL cholesterol ratio, valvular heart disease and prevalent MI. Body mass index, systolic blood pressure, hypertension treatment, diabetes, current smoking, total/HDL cholesterol ratio and valvular heart disease were modeled as time-dependent covariates.
Reference List


심부전 발생예측을 위한 새로운 접근법: 다중 표지자 사용 전략

강석민 교수 세브란스병원 심장내과

Summary

배경

조심실 좌심방과 심부전 발생과정에는 다양한 기전이 관여한다. 그러나 일반인들의 심부전 발생률 예측하는 데 있어 다양한 생물학적 지표들을 통한 다중 표지자 접근 전략의 유효성에 대해서는 아직 알려진 바가 없다.

방법 및 결과

Framingham Heart Study에 참여한 심부전 중상이 없는 2,754명(평균 연령 58세, 여성 54%)을 대상으로 6개의 생물학적 지표(C-reactive protein, plasminogen activator inhibitor-1, homocysteine, aldosterone to renin ratio, B-type natriuretic peptide and urinary albumin-to-creatinine ratio)를 이용해 심부전 발생을 예측하고자 하였다. 이러한 생물학적 지표들의 추가적인 유용성을 평가하기 위해 각 등급이 가진 기법을 사용하였다. 추적기간 동안(평균 9.4년) 95건의 심부전(남성 54건)이 발생하였다. 다변량분석 결과, 다중 표지자 접근 전략의 심부전 발생과 유의한 상관관계가 있다(OR=0.00005). 이 중에서 B-type natriuretic peptide(HR, 1.52; 95% CI, 1.24-1.87)와 urinary albumin-to-creatinine ratio(HR, 1.33; 95% CI, 1.11-1.66)가 심부전 발생을 예측하는 데 있어서 중요한 지표였다. 또한, 이 두 표지자는 심부전 발생 위험 예측 측면에 유의하게 증가하였으며(net reclassification improvement=0.13; P=0.002). 비해혈성 심부전 발생과도 유의하게 관련이 있었다.

결론

다중 표지자 사용 전략을 이용하여 B-type natriuretic peptide와 urinary albumin-to-creatinine ratio가 새로운 심부전 발생을 예측하는 중요한 위험인자임을 알 수 있었고 추가적인 유용성을 확인할 수 있었다.
심부전의 발생과 밀접한 관계를 보이는 다양한 기전들이 관여한다고 알려져 있으나, 아직까지 어떤 기전이 심부전 발생에 결정적인 역할을 하는지에 대해 정확하게 밝혀지지 않아. 또한, 이러한 기전을 대표하는 생물학적 지표들이 심부전과의 유의한 연관성을 보이고 있지만, 이와 관련된 연구가 아직 부족한 실정이다. 최근의 연구에서는 Framingham Heart Study에 참여한 일반인의 혈압을 통해 심부전 발생과 관련된 다양한 생물학적 지표들을 대상으로, 이들 중 aldosterone-to-renin ratio(renin-angiotensin-aldostrict system), C-reactive protein(inflammation), plasminogen activator inhibitor-1(fibrinolysis), B-type natriuretic peptide(natriuretic peptide system) homocysteine(oxidative stress)과 urinary albumin-to-creatinine ratio(endothelial function)의 대조군과 심부전의 새로운 발생을 예측하고자 하였다. 즉, 각각의 지표들을 여러 번 수측하는 것이 아니라 동 시에 여러 지표들을 조사하여 어떤 지표들이 심부전 발생에 중요한 영향을 미칠 수 있는지를 알아보기 위한 연구이다. 이 연구에서 B-type natriuretic peptide와 urinary albumin-to-creatinine ratio가 심부전 발생을 예측하는 중요한 지표들임을 알 수 있었다. 또한, 지표들 간의 관계를 분석하여 개별적으로 개별한 biomarker score에 따라 각 심부전 발생과 관련된 포지셔널 분석을 비롯하여 심부전 발생을 예측하고자 하였다. 심부전 발생에 있어 natriuretic peptide system의 발현이 endothelial dysfunction이 중요한 역할을 하는 것으로 보고하였다. 또한, 임상 수준의 기능을 가진 사람들의 임상상황을 관찰함으로써, 심부전 발생에 의한 심장 기능자의 비정상 상태가 심부전 발생에 기여할 수 있는가 하는 연구가 진행되어 왔다. 그러나 대상 환자의 약 50% 정도가 Framingham Risk Score-predicted 10-year CHD risk 10% 이상이었고, 실제 심부전 발생도 이 환자군의 약 80% 정도에서 발생한 사실을 고려한다면, 심혈관 위험도가 낮은 환자군에서 본 연구의 결과를 확대 적용하기에는 무리가 있다. 본 연구는 또한, 본 연구에서 제시한 biomarker score의 유용성을 여러 연구에서 검증이 필요하다고 생각되지만, 심부전 환자를 대상으로 연구를 수행하는 경우 유용할 수 있으며, 심부전 발생을 예측하는 데 있어 negative predictive value로서 중요한 역할을 할 수도 있다. 물론, 본 연구에서는 측정하지 않았지만 심부전 발생 기전에 관여하는 다른 생물학적 요인들에 대한 연구도 필요하다고 생각된다. 또한, 이러한 B-type natriuretic peptide와 urinary albumin-to-creatinine ratio의 대조자 문헌과 심부전 발병 관련 각종 생화학적 상태의 변화, 혈액내피 세포 기능과 변화 등과 연관성이 있는지에 대한 추가적인 연구가 필요하다고 생각된다. 본 연구 결과를 더욱 정당화해 보는 것이 필요하다.
Résumés d’articles

Valeur prédictive d’une série de marqueurs à l’égard du risque d’insuffisance cardiaque en milieu communautaire

Raghava S. Velagaleti, MD ; Philimon Gona, PhD ; Martin G. Larson, ScD ; Thomas J. Wang, MD ; Daniel Levy, MD ; Emelia J. Benjamin, MD, ScM ; Jacob Selhub, PhD ; Paul F. Jacques, ScD ; James B. Meigs, MD, MPH ; Geoffrey H. Tofler, MD ; Ramachandran S. Vasan, MD

Contexte—On sait que plusieurs voies biologiques sont activées lors du remodelage ventriculaire et de l’insuffisance cardiaque (IC) avérée. Aucune donnée n’est toutefois disponible sur la valeur additive d’un ensemble réduit de biomarqueurs (reflétant les voies impliquées dans la survenue de la maladie) en termes de prédiction du risque d’IC en milieu communautaire.

Méthodes et résultats—Nous avons recherché les liens unissant divers biomarqueurs à la survenue d’un premier épisode d’IC chez 2 754 participants à l’étude cardiologique de Framingham (âge moyen : 58 ans ; 54 % de femmes) qui étaient initialement indemnes d’une telle affection et chez lesquels six marqueurs biologiques avaient été systématiquement mesurés (à savoir la C-réactive protéine, l’inhibiteur I de l’activateur du plasminogène, l’homocystéine, le rapport aldostérone/rénine, le peptide natriurétique de type B et le rapport albuminurie/créatininurie). Nous avons estimé la statistique c, la calibration et l’indice d’amélioration de la reclassification nette du modèle pour apprécier la contribution additionnelle de chacun de ces marqueurs à la prédiction du risque. Nous avons également recherché les liens existant entre ces marqueurs et l’incidence des cas d’IC non ischémique chez les participants qui étaient indemnes de maladie coronaire à leur entrée dans l’étude. Au cours du suivi (durée moyenne : 9,4 ans), 95 premiers épisodes d’IC ont été enregistrés (dont 54 chez des hommes). Dans les modèles ajustés pour les variables multiples, notre série de biomarqueurs s’est montrée significativement corrélée avec le risque d’IC (p = 0,00005). A l’issue de la procédure d’éliminations rétrogardes successives, le peptide natriurétique de type B et le rapport albuminurie/créatininurie sont apparus comme des marqueurs prédicatifs majeurs du risque d’IC : les risques relatifs par augmentation de 1,66) Ces deux marqueurs ont significativement augmenté la statistique c des modèles standard de 0,84 (IC à 95 % : 0,80 à 0,88) à 0,86 (IC à 95 % : 0,83 à 0,90) ; ils ont également permis une meilleure reclassification des niveaux de risque (indice d’amélioration de la reclassification nette : 0,13 ; p = 0,002) et ont été corrélés de façon indépendante avec le risque d’IC non ischémique.

Conclusion—Par l’évaluation d’une série prédéfinie de marqueurs, nous avons pu établir que le peptide natriurétique de type B et le rapport albuminurie/créatininurie sont deux facteurs clés du risque de survenue d’une IC de novo et qui s’ajoutent à la valeur prédicative des facteurs de risque classiques. (Traduit de l’anglais : Multimarker Approach for the Prediction of Heart Failure Incidence in the Community. Circulation. 2010;122;1700–1706.)

Mots clés : biomarqueurs ■ épidémiologie ■ insuffisance cardiaque ■ peptides natriurétiques ■ risque ■ prédiction

Influence du lasofoxiène sur l’incidence des événements cardiovasculaires chez la femme ménopausée ostéoporotique

Résultats à cinq ans de l’essai Postmenopausal Evaluation and Risk Reduction With Lasofoxiène (PEARL)

Kristine Ensrud, MD, MPH ; Andrea LaCroix, PhD ; John R. Thompson, PhD ; David D. Thompson, PhD ; Richard Eastell, MD ; David M. Reid, MD ; Slobodan Vukicevic, MD ; Jane Cauley, DrPH ; Elizabeth Barrett-Connor, MD ; Roisin Armstrong, PhD ; Francine Welty, MD, PhD ; Steven Cummings, MD

Contexte—Dans l’essai PEARL (Postmenopausal Evaluation and Risk Reduction With Lasofoxiène [Evaluation du lasofoxiène dans la réduction des risques encourus après la ménopause]), les femmes traitées par le lasofoxiène à la posologie de 0,5 mg/jour ont présenté un moindre risque d’événement coronarien majeur et d’accident vasculaire cérébral (AVC), alors que celles ayant reçu le médicament à raison de 0,25 mg/jour ont été exposées à un plus faible risque d’AVC. En revanche, les deux doses de lasofoxiène ont majoré le risque d’événement thromboembolique veineux. Dans la présente publication, nous présentons des données complètes sur les événements cibles cardiovasculaires en examinant notamment chacun des