Journal of the American College of Cardiology Vol. 60, No. 14, 2012

Published by Elsevier Inc.
ISSN 0735-1097/$36.00
http://dx.doi.org/10.1016/j.jacc.2012.04.053

Biomarkers

Galectin-3, a Marker of Cardiac Fibrosis, Predicts Incident Heart Failure in the Community

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Objectives The aim of this study was to examine the relation of galectin-3 (Gal-3), a marker of cardiac fibrosis, with incident heart failure (HF) in the community.

Background Gal-3 is an emerging prognostic biomarker in HF, and experimental studies suggest that Gal-3 is an important mediator of cardiac fibrosis. Whether elevated Gal-3 concentrations precede the development of HF is unknown.

Methods Gal-3 concentrations were measured in 3,353 participants in the Framingham Offspring Cohort (mean age 59 years; 53% women). The relation of Gal-3 to incident HF was assessed using proportional hazards regression.

Results Gal-3 was associated with increased left ventricular mass in age-adjusted and sex-adjusted analyses ($p = 0.001$); this association was attenuated in multivariate analyses ($p = 0.06$). A total of 166 participants developed incident HF and 468 died during a mean follow-up period of 11.2 years. Gal-3 was associated with risk for incident HF (hazard ratio [HR]: 1.28 per 1 SD increase in log Gal-3; 95% confidence interval [CI]: 1.14 to 1.43; $p < 0.0001$) and remained significant after adjustment for clinical variables and B-type natriuretic peptide (HR: 1.23; 95% CI: 1.04 to 1.47; $p = 0.02$). Gal-3 was also associated with risk for all-cause mortality (multivariable-adjusted HR: 1.15; 95% CI: 1.04 to 1.28; $p = 0.01$). The addition of Gal-3 to clinical factors resulted in negligible changes to the C-statistic and minor improvements in net reclassification improvement.

Conclusions Higher concentration of Gal-3, a marker of cardiac fibrosis, is associated with increased risk for incident HF and mortality. Future studies evaluating the role of Gal-3 in cardiac remodeling may provide further insights into the role of Gal-3 in the pathophysiology of HF. (J Am Coll Cardiol 2012;60:1249–56) © 2012 by the American College of Cardiology Foundation

Heart failure (HF) accounts for more than 1 million hospital admissions per year, with an estimated cost exceeding $39 billion annually in the U.S. (1). The development of HF is often a clinically silent process, with progressive cardiac remodeling that eventually leads to symptomatic presentation late in the course of disease progression. After HF diagnosis, nearly 60% of men and 45% of women will die within 5 years (1). Although most therapies are implemented during the symptomatic phase of HF, when extensive remodeling has already occurred, strategies that target patients with cardiac remodeling before the onset of symptoms may prevent complications associated with HF (2,3). Cost-effective strategies to identify this subgroup of patients are of great interest, as outlined in the American College of Cardiology and American Heart Association guidelines (4).

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Although cardiac imaging of the general population is not recommended, a biomarker strategy to screen and identify patients to refer for diagnostic noninvasive cardiac imaging may be useful (5). This may facilitate the early recognition of asymptomatic left ventricular (LV) dysfunction and the initiation of therapy to favorably alter the course of progression to HF.

Cardiac fibrosis is an important contributor to the pathophysiology of LV systolic and diastolic dysfunction. It is also a pathologic phenomenon common to cardiac remodeling

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Manuscript received March 23, 2012; accepted April 18, 2012.

Abbreviations and Acronyms

BNP = B-type natriuretic peptide
CI = confidence interval
CKD = chronic kidney disease
eGFR = estimated glomerular filtration rate
Gal-3 = galectin-3
HF = heart failure
HR = hazard ratio
IDI = integrated discrimination improvement
LV = left ventricular
LVDD = left ventricular end-diastolic dimension
NRI = net reclassification improvement

caused by hypertensive, ischemic, and other conditions affecting the myocardium. Galectin-3 (Gal-3) is a beta-galactoside-binding lectin that appears to be a mediator of cardiac fibrosis in a number of recent experimental studies (6,7).

Gal-3 has been related to mortality in patients with acute and chronic HF (8–11), as well as in the general population (12). The role of Gal-3 as a predictor of incident HF in apparently healthy subjects has not been studied. We sought to examine the clinical correlates of Gal-3 to explore mechanisms by which Gal-3 may be associated with an adverse cardiovascular prognosis. We also examined the cross-sectional relations of Gal-3 to LV structure and function, to assess whether Gal-3 is associated with subclinical changes in cardiac function. Last, we sought to study the association of Gal-3 levels and incident HF events in the community. We hypothesized that Gal-3, a prognostic biomarker in patients with HF, would be associated with incident HF.

Methods

Participants. The Framingham Heart Study is a longitudinal community-based cohort initiated in 1948 to prospectively study cardiovascular disease and associated risk factors. The Framingham Offspring Cohort includes children (and spouses of children) of the original cohort participants, and participants have been examined approximately every 4 years since its inception in 1971 (13). Each examination includes routine questionnaires, physical examination, anthropometry, and blood testing. Gal-3 levels were measured at the sixth examination (1995 to 1998). Of 3,450 participants with sample available for Gal-3 measurement, a total of 97 participants were excluded because of prevalent HF (n = 40), stage IV kidney disease (n = 10), missing covariates (n = 39), missing Gal-3 measurements (n = 2), and extreme Gal-3 outliers (>5 log SDs above or below the log-transformed mean, n = 6, specified a priori), leaving 3,353 participants (97%) for analysis.

Biomarker measurement. Blood samples were collected after an overnight fast and immediately centrifuged and stored at −70°C until assayed. Plasma concentrations of Gal-3 were measured using an enzyme-linked immunosorbent assay (BG Medicine, Waltham, Massachusetts) (14). The lower detection limit was 1.32 ng/ml, with an upper detection limit of 96.6 ng/ml. Across this measurement range, the within-run and total precision are reported between 2.1% and 5.7% and 4.2% and 12.0%, respectively (14). B-type natriuretic peptide (BNP) was previously measured (15).

Clinical assessment. Participants underwent a comprehensive clinical assessment at the sixth Offspring Cohort examination (13). Hypertension was defined as a systolic blood pressure ≥140 mm Hg, a diastolic blood pressure ≥90 mm Hg, or current antihypertensive drug treatment. Cardiovascular events were adjudicated by a 3-physician panel after review of medical records. History of coronary heart disease included prior myocardial infarction, acute coronary insufficiency (prolonged ischemic symptoms with new electrocardiographic abnormalities in the absence of biomarker elevations indicative of infarction), or angina pectoris. Atrial fibrillation was determined after examining all available electrocardiograms. Valvular heart disease was defined as a systolic murmur of grade 3/6 or higher or any diastolic murmur. Total and high-density lipoprotein cholesterol levels were measured. Diabetes mellitus was defined as a fasting glucose level ≥126 mg/dl, nonfasting glucose ≥200 mg/dl, or the use of insulin or oral hypoglycemic medications. Estimated glomerular filtration rate (eGFR) was calculated using the Modification of Diet in Renal Disease equation (16).

Definition of HF. At each follow-up examination or health update, interim cardiovascular disease events were identified and medical records obtained. Initial HF was confirmed by a panel of 3 physicians after systematic review of outpatient and hospital records using established protocols and Framingham criteria (17). The present study included initial HF events occurring between the baseline examination (1995 to 1998) through the end of 2008 as incident events.

Echocardiographic methods. A total of 2,425 participants with Gal-3 measurements also underwent routine M-mode and 2-dimensional echocardiography (18) and had complete data for analysis. LV end-diastolic dimension (LVDD), LV end-systolic dimension, left atrial end-systolic diameter, and end-diastolic LV septal and posterior wall thicknesses were measured according to American Society of Echocardiography guidelines (19). Fractional shortening was calculated as: [(LVDD − LV end-systolic dimension)/LVDD] × 100, and LV systolic dysfunction was defined as fractional shortening <29%. LV mass was calculated as: $0.8\{1.04[(\text{LVDD} + \text{LV posterior wall thickness} + \text{LV septal wall thickness})^3 - \text{LVDD}^3]\} + 0.6$ (20). LV mass was indexed to height$^{2.7}$ (21), and elevated LV mass was defined as an indexed value greater than or equal to the sex-specific 80th percentile.

Statistical analysis. Because of non-normality, Gal-3, eGFR, and BNP were log transformed for subsequent analyses. Baseline clinical characteristics were summarized by sex-specific quartiles of log Gal-3 and trends in means across quartiles examined. Multivariate regression analysis of Gal-3 correlates was performed using a stepwise selection model with inclusion of variables at $p < 0.05$. Because of sex differences in Gal-3 distribution, sex-standardized log Gal-3 was used for correlation and regression analyses.

The associations of Gal-3 with measures of cardiac structure and function (including LV mass, left atrial end-systolic diameter, and LV fractional shortening) were examined using linear regression models adjusting for: 1) age, sex, and height; and 2) age, sex, height, systolic blood pressure, antihypertensive medication use, diabetes mellitus, and previous myocardial infarction. Analyses of fractional shortening were not adjusted for height, and analyses of left atrial end-systolic diameter were also adjusted for valvular heart disease. Participants with prevalent atrial fibrillation were excluded from echocardiographic analyses.

Crude HF incidence rates were estimated by sex-specific Gal-3 quartile. The cumulative incidence of HF across Gal-3 quartiles was examined using a Kaplan-Meier-like method while accounting for competing risk for death (22). Multivariable Cox proportional hazards regression models were constructed to evaluate the association of Gal-3 with incident HF events and with all-cause mortality (23). Models were created adjusting for 1) age and sex; 2) age, sex, systolic blood pressure, antihypertensive medication use, body mass index, diabetes mellitus, current smoking status, prevalent coronary heart disease, valvular heart disease, and atrial fibrillation; and 3) all covariates in model 2 plus BNP. Proportional hazards assumptions were met. Analyses for mortality were further adjusted for eGFR and total and high-density lipoprotein cholesterol. In secondary analyses, we examined the association of Gal-3 and cardiovascular death (defined as death from coronary heart disease, cerebrovascular disease, or other cardiovascular cause), adjusting for the same covariates as analyses for all-cause mortality.

We adjusted HF analyses for eGFR in secondary analyses, because changes in kidney function may mediate Gal-3 effects on incident HF. We conducted sensitivity analyses, excluding 283 patients with prevalent chronic kidney disease (CKD; defined as eGFR <60 ml/min/1.73 m^2), and we used a time-dependent covariate to adjust for incident CKD events in analyses for both incident HF and mortality.

To assess the incremental benefit of Gal-3 in the prediction of HF and mortality risk, C-statistics were compared between models with traditional risk factors with and without Gal-3 (24). We estimated the integrated discrimination improvement (IDI) and the category-free net reclassification improvement (NRI) metric for the addition of Gal-3 in fully adjusted models (25,26). All statistical analyses were conducted using SAS version 9.2 for Windows (SAS Institute Inc., Cary, North Carolina).

Results

The baseline clinical characteristics of 3,353 participants are displayed by Gal-3 quartiles in Table 1. The mean age was 59 years, and 53% of participants were women. The distribution of Gal-3 levels in our sample is shown in Online Figure 1. Gal-3 concentrations were higher in women compared with men ($p < 0.05$), with a median Gal-3 level in women of 14.3 ng/ml (interquartile range: 12.0 to 16.8 ng/ml) versus 13.1 ng/ml (interquartile range: 11.1 to 15.4 ng/ml) in men. Participants with higher Gal-3 levels were older and had a higher prevalence of traditional cardiovascular risk factors, including hypertension, diabetes mellitus,

Table 1 Baseline Characteristics of 3,353 Participants by Sex-Specific Gal-3 Quartile

Characteristic	Quartile of Gal-3*				p for Trend
	1 (n = 835)	2 (n = 842)	3 (n = 842)	4 (n = 834)	
Clinical					
Age (yrs)	55 ± 9	58 ± 9	60 ± 9	64 ± 10	<0.0001
Women	454 (54%)	431 (51%)	436 (52%)	461 (55%)	0.98
Systolic blood pressure (mm Hg)	124 ± 18	127 ± 18	130 ± 19	132 ± 20	<0.0001
Diastolic blood pressure (mm Hg)	75 ± 9	76 ± 9	76 ± 9	75 ± 10	0.34
Antihypertensive medication use	134 (16%)	193 (23%)	250 (30%)	355 (43%)	<0.0001
Diabetes mellitus	56 (7%)	66 (8%)	81 (10%)	115 (14%)	<0.0001
Coronary heart disease	32 (4%)	40 (5%)	66 (8%)	106 (13%)	<0.0001
Body mass index (kg/m^2)	26.9 (4.7%)	27.6 (5.1%)	28.5 (5.2%)	28.6 (5.3%)	<0.0001
Smoking	121 (14%)	132 (16%)	149 (18%)	109 (13%)	0.63
Laboratory					
Total cholesterol (mg/dl)	200 ± 36	205 ± 37	209 ± 37	208 ± 42	<0.0001
HDL cholesterol (mg/dl)	54 ± 17	52 ± 16	51 ± 16	48 ± 15	<0.0001
eGFR (ml/min/1.73 m^2)	94 ± 22	90 ± 24	88 ± 25	80 ± 25	<0.0001
BNP (pg/ml)	12.8 ± 15.6	13.7 ± 17.7	14.7 ± 18.3	22.4 ± 30.6	<0.0001
Echocardiography (n = 2,425)*					
Left ventricular mass (g/m^2)	156 ± 42	159 ± 44	163 ± 45	166 ± 45	<0.0001
Fractional shortening (%)	37.1 ± 5.2	37.1 ± 5.7	37.1 ± 5.4	37.3 ± 6.2	0.59
Left atrial dimension (mm)	39.0 ± 5.0	39.3 ± 5.2	39.3 ± 5.1	40.0 ± 5.4	0.001

Values are mean ± SD or n (%). *Lower and upper limits for Gal-3 quartiles for men and women were as follows: men: quartile 1, 3.9 to 11.1 ng/ml; quartile 2, 11.1 to 13.1 ng/ml; quartile 3, 13.1 to 15.4 ng/ml; and quartile 4, 15.4 to 47.7 ng/ml; women: quartile 1, 5.0 to 12.0 ng/ml; quartile 2, 12.0 to 14.3 ng/ml; quartile 3, 14.3 to 16.8 ng/ml; and quartile 4, 16.8 to 52.1 ng/ml.

BNP = B-type natriuretic peptide; eGFR = estimated glomerular filtration rate; Gal-3 = galectin-3; HDL = high-density lipoprotein.

Table 2 Clinical Correlates of Gal-3 in 3,353 Participants

Correlate	Coefficient	Standard Error	p Value
Age	0.222	0.019	<0.0001
Antihypertensive treatment	0.180	0.039	<0.0001
Body mass index	0.113	0.016	<0.0001
Coronary heart disease	0.168	0.065	0.009
eGFR	−0.141	0.017	<0.0001
BNP	0.041	0.018	0.02

Log Gal-3 was standardized by sex. The regression coefficients indicate the increase in log Gal-3 in the presence versus absence of the trait for dichotomous variables and per 1 standard deviation increase for continuous variables (per 10-year increase in age, per 5.1 kg/m^2 increase in body mass index, per 0.26 increase in log eGFR, and per 0.90 increase in log BNP). The following variables were not significant in the stepwise selection model ($p > 0.05$): systolic blood pressure, diabetes, smoking, total and HDL cholesterol, valvular heart disease, and atrial fibrillation.

Abbreviations as in Table 1.

previous coronary heart disease, higher body mass index, and lower eGFR (p for trend <0.0001 for all).

Clinical correlates of Gal-3. In multivariable analyses, Gal-3 was positively associated with age, hypertension, body mass index, prevalent coronary heart disease, and BNP, and negatively associated with eGFR (Table 2). The R^2 value of this model was 0.15. There was a weak correlation between Gal-3 and BNP (age- and sex-adjusted Pearson partial correlation, r = 0.05, p = 0.002).

Among 2,425 participants who had usable echocardiographic data, a 1 standard deviation increase in log Gal-3 was associated with 2-fold increased odds of having elevated LV mass in age- and sex-adjusted analyses (95% confidence interval [CI]: 1.33 to 3.08; p = 0.001). When LV mass was used as a continuous variable, higher Gal-3 remained positively associated with higher LV mass (p = 0.03). This association was attenuated after adjustment for clinical covariates (Table 3). Gal-3 was not associated with fractional shortening, LV systolic dysfunction, or left atrial size.

Gal-3 and incident HF events. During a mean follow-up period of 11.2 years, 166 patients (5.1%) experienced first HF events. The crude HF incidence rate increased over Gal-3 quartiles, with rates of 2.8, 3.8, 5.2, and 12.4 events per 1,000 person-years in quartiles 1 through 4, respectively. Figure 1 demonstrates higher cumulative incidence of HF with increasing Gal-3 quartiles (log-rank test $p < 0.0001$). In age- and sex-adjusted analyses, a 1 standard deviation increase in log Gal-3 was associated with a 28% increased risk for incident HF (95% CI: 1.14 to 1.43, $p < 0.0001$) (Table 4). After multivariable adjustment and the addition of BNP, Gal-3 remained predictive of HF risk (hazard ratio [HR]: 1.23 per standard deviation increment in log Gal-3; 95% CI: 1.04 to 1.47; p = 0.02). BNP in the same model was associated with a 46% increased risk for HF (95% CI: 1.23 to 1.75; $p < 0.0001$). In analyses examining the association of Gal-3 quartile and incident HF, there was a significant increase in HF risk across quartiles in age- and sex-adjusted analyses (p = 0.004), but this did not reach statistical significance after multivariable adjustment (p = 0.11) (Online Table 1).

In secondary analyses, adjusting for eGFR had modest impact (multivariable-adjusted HR: 1.22; 95% CI: 1.01 to 1.46; p = 0.04; multivariable-adjusted and BNP-adjusted HR: 1.19; 95% CI: 0.99 to 1.42; p = 0.06). Sensitivity analyses excluding patients with prevalent CKD or adjusting for the development of incident CKD attenuated the association of Gal-3 and incident HF ($p > 0.05$ for both).

Gal-3 and mortality. There were 468 deaths during the follow-up period. Increasing Gal-3 quartiles were associated with higher all-cause mortality, as displayed in the cumulative incidence graphs in Figure 2. In age- and sex-adjusted analyses, a 1 standard deviation increase in log Gal-3 was associated with 24% increased risk for mortality (95% CI: 1.12 to 1.38; $p < 0.0001$) (Table 4). This association remained significant after accounting for clinical covariates and BNP (HR: 1.14; 95% CI: 1.02 to 1.27; p = 0.02). BNP was associated with a similar risk for mortality in the same model (HR: 1.12; 95% CI: 1.01 to 1.24; p = 0.03). The risk for mortality increased across Gal-3 quartiles (p for trend = 0.0007), and the fourth quartile was associated with >60% increased hazards of mortality compared with the first quartile (multivariable-adjusted and BNP-adjusted HR: 1.62; 95% CI: 1.18 to 2.22; p = 0.003) (Online Table 1).

In secondary analyses, Gal-3 was associated with cardiovascular death (98 events) in age- and sex-adjusted analyses (HR: 1.48; 95% CI: 1.21 to 1.82; p = 0.0002). This association was partly attenuated after adjustment for clinical covariates (multivariable-adjusted HR: 1.25; 95% CI:

Table 3 Gal-3 Associations With Echocardiographic Traits in 2,425 Participants

Variable	Model 1*	p Value	Model 2†	p Value
Dichotomous variable				
Increased LV mass	2.02 (1.33–3.08)	0.001	1.54 (0.99–2.38)	0.06
LV systolic dysfunction	2.15 (0.99–4.65)	0.05	1.72 (0.75–3.93)	0.20
Continuous variables				
LV mass	6.440 (2.880)	0.03	1.700 (2.804)	0.54
Fractional shortening	−0.007 (0.005)	0.14	−0.007 (0.004)	0.14
Left atrial dimension	−0.004 (0.038)	0.91	−0.062 (0.037)	0.09

Values are odds ratio (95% confidence interval) or coefficient (standard error). Odds ratios and regression coefficients denote change associated with a 1 SD increase in log Gal-3 level. *Adjusted for age and sex. Analyses for LV mass and left atrial dimension were also adjusted for height. †Adjusted for age, sex, diabetes, systolic blood pressure, antihypertensive medication, and previous myocardial infarction. Additionally, analyses for LV mass and left atrial dimension were adjusted for height, and left atrial dimension analyses were adjusted for valvular heart disease.

Gal-3 = galectin-3; LV = left ventricular.

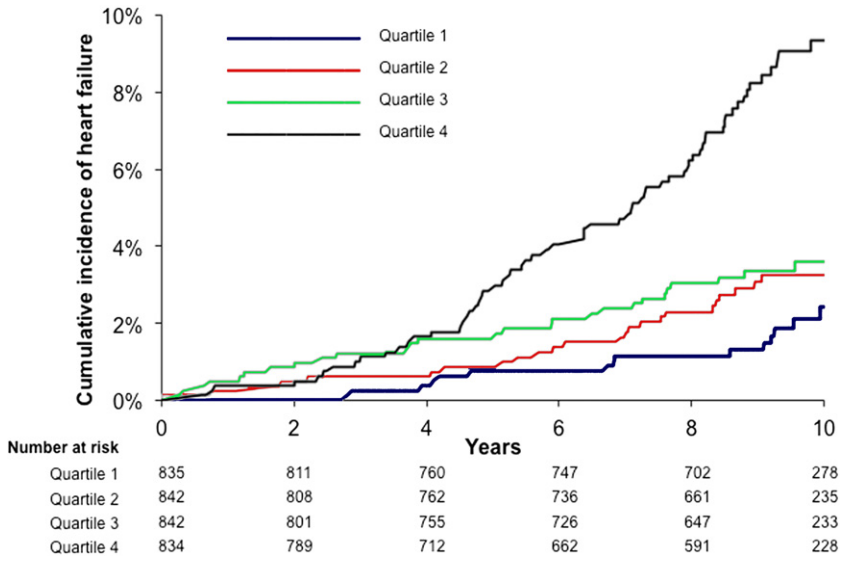

Figure 1 HF and Gal-3 Quartiles

The cumulative incidence of heart failure (HF) increased with higher galectin-3 (Gal-3) quartiles.

1.01 to 1.54; p = 0.045; multivariable-adjusted and BNP-adjusted HR: 1.21; 95% CI: 0.98 to 1.49; p = 0.08).

The exclusion of individuals with prevalent CKD did not attenuate the association of Gal-3 with mortality (multivariable-adjusted HR: 1.21; 95% CI: 1.08 to 1.36; p = 0.0009), and the association persisted after further adjusting for incident CKD (HR: 1.25; 95% CI: 1.10 to 1.43; p = 0.0008).

Performance of Gal-3 as a biomarker. When added to the clinical model for HF, Gal-3 did not substantially increase the C-statistic (0.855 to 0.859), with similar findings in the prediction of all-cause mortality (Table 5). Improvements in the IDI and relative IDI were small and comparable to the addition of BNP alone into the clinical model for mortality. The category-free NRI for the addition of Gal-3 in predicting HF was 0.20 (95% CI: 0.02 to 0.40), representing a weak effect size. Similar magnitudes were observed for the category-free NRI in the prediction of all-cause mortality.

Gal-3 in HF with preserved versus reduced ejection fraction. Of 166 participants with incident HF events, 140 (84%) underwent assessment of LV function at or around the time of HF onset. Of these, 63 were classified as having HF with preserved ejection fraction and 77 as having HF with reduced ejection fraction. There was no difference in baseline Gal-3 levels in participants who developed HF with preserved versus reduced ejection fraction (16.3 ± 4.5 ng/ml vs. 15.8 ± 4.2 ng/ml, respectively, p = 0.54).

Table 4 Association of Plasma Gal-3 Levels, Incident HF, and Mortality

Outcome	Model	HR (95% CI)	p Value
Incident HF			
	Age and sex adjusted	1.39 (1.17–1.65)	0.0002
	Multivariable adjusted*	1.27 (1.06–1.52)	0.01
	Multivariable adjusted + BNP	1.23 (1.04–1.47)	0.02
All-cause mortality			
	Age and sex adjusted	1.25 (1.12–1.39)	<0.0001
	Multivariable adjusted*	1.15 (1.04–1.29)	0.01
	Multivariable adjusted + BNP	1.14 (1.03–1.28)	0.02

Hazard ratios (HRs) and 95% confidence intervals (CIs) denote hazard associated with a 1 SD increase in log Gal-3 levels. This increase is equivalent to comparing a Gal-3 level of 14.0 ng/ml (sample mean) with a level of 18.1 ng/ml. *Adjusted for age, sex, systolic blood pressure, antihypertensive treatment, body mass index, diabetes mellitus, smoking, prevalent coronary heart disease, atrial fibrillation, and valvular heart disease. Mortality analyses were additionally adjusted for eGFR and total and HDL.

HF = heart failure; other abbreviations as in Table 1.

Discussion

Our findings demonstrate that higher levels of Gal-3, a marker of cardiac fibrosis, are associated with an increased risk for incident HF and all-cause mortality in the community. Previous studies have examined the prognostic value of Gal-3 in patients with existing HF. To our knowledge, our study is the first to report the association of Gal-3 with risk for new-onset HF in apparently healthy subjects. Our data also suggest that the association of Gal-3 with incident HF may be influenced by kidney function. Further studies on the link between Gal-3, kidney function, and myocardial injury and fibrosis will help elucidate the potential role of Gal-3 in the pathophysiology of HF.

Gal-3 is emerging as a prognostic biomarker in patients with HF (8–11,27,28). More recently, higher Gal-3 levels were found to be associated with all-cause mortality in a community-based cohort (12). Our findings substantiate the prognostic role of Gal-3 with respect to all-cause mortality. Gal-3 is an indicator not only of myocardial

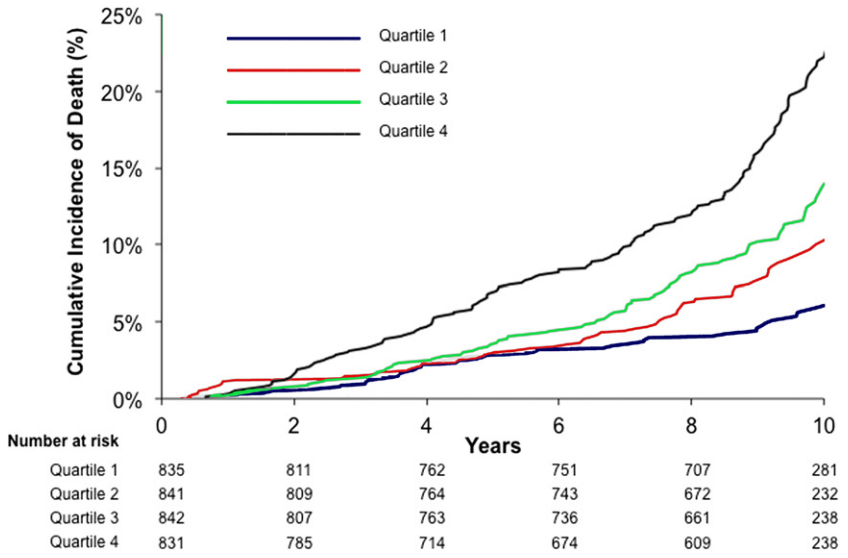

Figure 2 Mortality and Gal-3 Quartiles

The cumulative incidence of all-cause mortality increased with higher galectin-3 (Gal-3) quartiles.

fibrosis but also other fibrotic conditions, including liver cirrhosis (29,30) and pulmonary fibrosis (31), all of which could increase the risk for overall mortality. Beyond the association with all-cause mortality, a recent case-control study demonstrated an association of Gal-3 with HF risk after acute coronary syndromes (32). Ours is the first study to extend these findings to a longitudinal cohort of ostensibly healthy subjects and to demonstrate the role of Gal-3 as a predictor of new-onset HF in the community.

Experimental evidence suggests that Gal-3 may be a mediator of fibrosis (33). Gal-3 is up-regulated in a number of human fibrotic disease entities, including liver cirrhosis (29,30) and pulmonary fibrosis (31). Gal-3$^{-/-}$ mice are protected against hepatic and renal fibrosis, and Gal-3 appears to be required for transforming growth factor–beta–mediated myofibroblast activation and matrix production (30,34). Gal-3 is the most overexpressed gene in transgenic Ren-2 rats that rapidly progress to HF (6). Gal-3 is expressed in activated macrophages, with binding sites localized to the myocardial extracellular matrix and cardiac fibroblasts, where it induces fibroblast proliferation, collagen deposition, and ventricular dysfunction (6). Infusion of Gal-3 into the pericardial space leads to cardiac dysfunction in rats, a process that appears to be mediated via the transforming growth factor–beta/Smad3 signaling pathway (7). In clinical studies, Gal-3 is correlated with markers of extracellular matrix turnover, supporting its role in collagen metabolism (35).

This collective experimental evidence suggests that Gal-3 may play a causal in cardiac remodeling. Although the incremental prognostic value of adding Gal-3 to existing clinical risk factors, particularly above and beyond BNP, was

Table 5 Performance Metrics of Gal-3 in Risk Prediction Models

	C-Statistic (95% CI)	IDI (95% CI)	Relative IDI (95% CI)	Category-Free NRI (95% CI)
Incident HF				
Clinical model*	0.855 (0.823 to 0.887)			
Clinical model + Gal-3	0.859 (0.828 to 0.890)	0.001 (–0.002 to 0.005)	0.014 (–0.022 to 0.052)	0.203 (0.018 to 0.397)
Clinical model + BNP	0.869 (0.839 to 0.898)	0.007 (–0.002 to 0.017)	0.070 (–0.021 to 0.164)	0.290 (0.110 to 0.473)
Clinical model + BNP + Gal-3†	0.871 (0.842 to 0.900)	0.001 (–0.002 to 0.005)	0.011 (–0.023 to 0.044)	0.162 (–0.028 to 0.360)
All-cause mortality				
Clinical model*	0.785 (0.762 to 0.808)			
Clinical model + Gal-3	0.786 (0.763 to 0.809)	0.001 (–0.001 to 0.004)	0.007 (–0.008 to 0.021)	0.184 (0.066 to 0.297)
Clinical model + BNP	0.785 (0.762 to 0.808)	0.002 (–0.001 to 0.004)	0.009 (–0.004 to 0.022)	0.108 (–0.011 to 0.226)
Clinical model + BNP + Gal-3†	0.786 (0.763 to 0.809)	0.001 (–0.002 to 0.003)	0.004 (–0.010 to 0.018)	0.178 (0.066 to 0.291)

*Clinical model includes age, sex, systolic blood pressure, antihypertensive treatment, body mass index, diabetes mellitus, smoking, prevalent coronary heart disease, atrial fibrillation, and valvular heart disease. Mortality analyses were additionally adjusted for prevalent HF, eGFR, and total and HDL cholesterol. †IDI, relative IDI, and category-free NRI represented are for the addition of Gal-3 to the clinical model or to the clinical model plus BNP.

IDI = integrated discrimination improvement; NRI = net reclassification improvement; other abbreviations as in Tables 1, 3, and 4.

marginal, the potential clinical role of Gal-3 may be pathobiological rather than prognostic in nature. Our findings are consistent with the notion that active fibrosis may precede clinical manifestations of HF by many years. Although most HF therapies are initiated late in the course of the disease, the identification of fibrosis before impairment of LV function may offer a window of opportunity to initiate targeted preventive treatment early in the course of the disease. Because of its putative role as a mediator of fibrosis, directly modulating the Gal-3 pathway may be beneficial. Gal-3 levels can be modulated with modified citrus pectin, a soluble dietary fiber found in citrus fruit (36). This pectin derivative can bind to the carbohydrate recognition domain of Gal-3, altering its bioactivity (37). Treatment with modified citrus pectin decreased Gal-3 expression and significantly attenuated renal fibrosis and inflammation in an animal model of acute kidney injury (38), and future studies examining the effect of Gal-3 inhibition on cardiovascular end points would be of high interest.

Notably, we observed a strong association of Gal-3 with kidney dysfunction, a finding that has been corroborated by previous studies in patients with and without existing HF (10,12,28,39). We also found that adjusting for kidney function attenuated the association of Gal-3 with incident HF events. This is consistent with prior studies in HF, in which adjustment for kidney function appeared to attenuate in part the prognostic impact of Gal-3 (39) and in which the association of Gal-3 with kidney function appeared to overshadow associations with cardiac structure and function (28). It may be that worsening kidney function mediates Gal-3 effects on cardiac remodeling and HF. Alternatively, elevated levels of Gal-3 in both the kidney and the heart may lead to HF, and consequently adjustment for kidney function may obscure the association of Gal-3 with HF risk.

We found that Gal-3 was associated with elevated LV mass in age-adjusted and sex-adjusted analysis, although this relation was only marginally significant in multivariable analyses. Gal-3 may be a stronger prognostic marker in those with preserved compared with reduced ejection fractions (39), and Gal-3 levels have been associated with measures of diastolic function in patients presenting with acute decompensated HF (11). Biologically, it is plausible that a matrix and fibrosis marker such as Gal-3 may play a more prominent role in those with HF with preserved ejection fraction. However, in exploratory analyses, we found no difference in baseline Gal-3 concentrations in participants who eventually developed HF with preserved versus reduced ejection fraction.

Study limitations. Although the addition of Gal-3 to clinical factors resulted in improved classification as assessed by the category-free NRI, changes in the C-statistic and IDI were negligible. It may be that Gal-3 in combination with other novel biomarkers in a multimarker approach might be useful, and the comparison of Gal-3 with other emerging biomarkers of HF, such as high-sensitivity troponin, N-terminal pro-BNP, and soluble ST2 will need to be explored in future studies. The number of HF events was modest and likely limited our power to conduct quartile analyses or other more complex analyses examining the role of kidney function in the association of Gal-3 and HF. In addition to its role in fibrosis in several organ systems, Gal-3 has also been associated with tumorigenesis in thyroid cancer and other malignancies (40). Circulating Gal-3 levels have not been elevated in these conditions (41), but we cannot exclude the possibility that Gal-3 might act in several pathophysiologic pathways to increase mortality risk. Although secondary analyses demonstrate a suggestive association with cardiovascular death, Gal-3 may still reflect noncardiac processes. Further elucidation of Gal-3 in relation to cardiac remodeling, including more sensitive measures of diastolic function or direct measures of cardiac fibrosis would be of great interest in future studies. Last, our study was limited to a predominantly white study sample, limiting generalization to other populations.

Conclusions

Higher circulating Gal-3 concentrations are associated with increased risk for new-onset HF and all-cause mortality in the community. Future potential clinical uses of Gal-3 measurement might include the identification of asymptomatic subjects with early evidence of cardiac fibrosis, in whom targeted therapies may be useful to delay the onset of HF. Animal data suggest that Gal-3 is a mediator of fibrosis, and directly targeting the Gal-3 pathway may represent a future preventive treatment strategy.

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Key Words: biomarker ■ epidemiology ■ heart failure ■ prognosis.

APPENDIX

For a supplementary table and figure, please see the online version of this article.