









Cardiovascular biomarkers predict post-discharge re-hospitalization risk and mortality among Swedish heart failure patients

John Molvin^{1,2*} , Amra Jujic^{1,2} , Erasmus Bachus^{1,3} , Widet Gallo¹ , Gordana Tasevska-Dinevska² , Hannes Holm¹, Olle Melander^{1,3} , Artur Fedorowski^{1,2}  and Martin Magnusson^{1,2,4} 

¹Department of Clinical Sciences, Lund University, Malmö, Malmö, Sweden; ²Department of Cardiology, Skåne University Hospital, Malmö, Carl-Bertil Laurells gata 9, 214 28, Malmö, Sweden; ³Department of Internal Medicine, Skåne University Hospital, Malmö, Malmö, Sweden; ⁴Wallenberg Centre for Molecular Medicine, Lund University, Lund, Lund, Sweden

Aim The aim of this study was to assess the predictive role of biomarkers, associated with cardiovascular stress and its neuroendocrine response as well as renal function, in relation to mortality and risk of re-hospitalization among consecutive patients admitted because of heart failure (HF).

Methods and results A total of 286 patients (mean age, 75 years; 29% women) hospitalized for newly diagnosed or exacerbated HF were analysed. Associations between circulating levels of mid-regional pro-adrenomedullin (MR-proADM), copeptin, C-terminal pro-endothelin-1, N-terminal pro-brain natriuretic peptide (NT-proBNP), cystatin C, and all-cause mortality as well as risk of re-hospitalization due to cardiac causes were assessed using multivariable Cox regression models. A two-sided Bonferroni-corrected P -value of $0.05/5 = 0.010$ was considered statistically significant. All biomarkers were related to echocardiographic measurements of cardiac dimensions and function. A total of 57 patients died (median follow-up time, 17 months). In the multivariable-adjusted Cox regression analyses, all biomarkers, except C-terminal pro-endothelin-1, were significantly associated with increased mortality: NT-proBNP [hazard ratio (HR) 1.85, 95% confidence interval (CI) 1.17–2.17; $P = 4.0 \times 10^{-4}$], MR-proADM (HR 1.94, 95% CI 1.36–2.75; $P = 2.2 \times 10^{-4}$), copeptin (HR 1.70, 95% CI 1.22–2.36; $P = 0.002$), and cystatin C (HR 2.11, 95% CI 1.56–2.86; $P = 1.0 \times 10^{-6}$). A total of 90 patients were re-hospitalized (median time to re-hospitalization, 5 months). In multivariable Cox regression analyses, NT-proBNP was the only biomarker that showed significant association with risk of re-hospitalization due to cardiac causes (HR 1.43, 95% CI 1.10–1.87; $P = 0.009$).

Conclusions Among patients hospitalized for HF, elevated plasma levels of NT-proBNP, MR-proADM, copeptin, and cystatin C are associated with higher mortality after discharge, whereas NT-proBNP is the only biomarker that predicts the risk of re-hospitalization due to cardiac causes.

Keywords Heart failure (HF); Mid-regional pro-adrenomedullin (MR-proADM); Copeptin; C-terminal pro-endothelin-1 (CT-pro-ET-1); N-terminal pro-brain natriuretic peptide (NT-proBNP); Cystatin C

Received: 23 April 2019; Accepted: 1 June 2019

*Correspondence to: John Molvin, Department of Cardiology, Skåne University Hospital, Malmö, Carl-Bertil Laurells gata 9, 214 28 Malmö, Sweden. Email: john.molvin@med.lu.se

Introduction

Heart failure (HF) is not only one of our deadliest and most widespread diseases but also one of the most common causes of hospitalization and re-hospitalization.¹ Although the care of patients with HF has improved over the last decades, physicians need better tools to predict adverse events and risk stratify patients hospitalized for HF.

Biomarkers have been shown to have limited value for clinical assessment in addition to traditional risk factors in regard to prediction of cardiovascular disease.² However, biomarkers related to inflammation and haemodynamic stress have recently been shown to predict or rule out early post-discharge events in patients hospitalized for acute HF.³ In particular, creatinine, brain natriuretic peptides (BNPs), pro-adrenomedullin, and endothelin 1 (ET-1) were all significantly

higher in subjects that died because of HF. Furthermore, these four biomarkers also showed additive value in low-risk vs. high-risk prediction of early post-discharge death or HF re-admission in patients hospitalized for acute HF.³

In this study, we analysed the following biomarkers: mid-regional pro-adrenomedullin (MR-proADM), copeptin, C-terminal pro-endothelin-1 (CT-pro-ET-1), N-terminal pro-brain natriuretic peptide (NT-proBNP), and cystatin C; these are biomarkers associated with cardiovascular stress and the neuroendocrine response it incites as well as renal function to assess their predictive role in relation to mortality and risk of re-hospitalization in a Swedish prospective HF cohort. Finally, because echocardiography is the most common modality to diagnose and grade severity of HF, the plasma levels of the biomarkers were related to echocardiographic measurements of cardiac dimension and function.

Methods

Study population

The HeART and Brain Failure inVESTigation project in Malmö, Sweden (HARVEST—Malmö), is an ongoing study undertaken in patients hospitalized for HF (ICD-10: I50-) in Skåne University Hospital, Malmö.^{4,5} The inclusion criteria for the HARVEST study are admission to the Department of Cardiology or Internal Medicine for treatment of newly diagnosed or exacerbated HF. The only exclusion criterion is the inability to give informed consent. In case of severe cognitive impairment, informed consent has been collected from relatives.

Between March 2014 and October 2017, a total of 283 consecutive patients hospitalized for HF were included and underwent clinical examination. Of these, 268 patients had complete dataset on all covariates and were included in the present analysis. The study was approved by the ethical review board at Lund University, Sweden. A written informed consent was obtained from all participants.

Clinical examination

After admission to the clinical ward, study participants were examined with anthropometric measurements, and blood samples were drawn after overnight fast. Body mass index was calculated as kg/m², and data regarding the study participants' medication were collected. Prevalent diabetes was defined as either self-reported diagnosis of type 2 diabetes or use of antidiabetic medication. Hypertension was defined as either systolic blood pressure ≥ 140 mmHg or diastolic blood pressure ≥ 90 mmHg.⁶ A diagnosis of atrial fibrillation was based on previous hospital records or on admission electrocardiography. Information about patients' medication was retrieved at discharge.

Laboratory assays

Analyses of high-density lipoprotein and plasma cholesterol were carried out upon admission at the Department of Clinical Chemistry, Skåne University Hospital, Malmö, participating in a national standardization and quality control system. For the biomarker analyses, blood samples were collected after admission within 24 h in a fasting condition. Blood samples were stored at -80 °C.

N-terminal pro-brain natriuretic peptide

N-terminal pro-brain natriuretic peptide was analysed at the Department of Clinical Chemistry, Skåne University Hospital, Malmö, participating in a national standardization and quality control system using ElectroChemiluminescenceImmunoassay (Cobas NPU21571).

Cystatin C

Cystatin C was analysed at the Department of Clinical Chemistry, Skåne University Hospital, using an automated particle-based immunoassay (Hitachi Modular P analysis system; Roche, Basel, Switzerland).

Copeptin

Copeptin was measured at baseline using an ultrasensitive assay on KRYPTOR Compact Plus analyzers and a commercial sandwich immunoluminometric assay (Thermo Fisher Scientific, B.R.A.H.M.S Biomarkers) as previously described.⁷ The lower detection limit was 0.4 pmol/L, and the functional assay sensitivity (<20% inter-assay coefficient of variation) was less than 1 pmol/L.

Mid-regional pro-adrenomedullin

The MR-proADM levels were analysed at baseline via specific sandwich immunoluminometric assays (KRYPTOR, B.R.A.H.M.S, Berlin, Germany) in EDTA-treated plasma.⁸ Mean inter-assay coefficient of variation was <10%.⁹

C-terminal pro-endothelin-1

C-terminal pro-endothelin-1 was measured at baseline using Thermo Fisher Scientific B.R.A.H.M.S CT-pro-ET-1 KRYPTOR. The analytical detection limit of CT-pro-ET-1 was 0.4 pmol/L, and inter-laboratory coefficient of variation was <10% for values >10 pmol/L.¹⁰

Echocardiography

Conventional transthoracic echocardiograms were obtained using a Philips IE33 (Philips, Andover, MA, USA) with a 1–5 MHz transducer (S5-1) or with a GE Vingmed Vivid 7 Ultrasound (GE, Vingmed Ultrasound, Horten, Norway) with a 1–4 MHz transducer (M3S). All studies were performed by experienced sonographers. Cine loops were obtained from standard views (parasternal long axis and apical four chamber

and two chamber). Measurements were performed offline using Xcelera 4.1.1 (Philips Medical Systems, The Netherlands) according to the recommendations of the American Society of Echocardiography.¹¹ Internal left and right ventricular dimensions were measured from parasternal long-axis view at end diastole. Measurements of wall thickness were obtained in two-dimensional end-diastolic parasternal long-axis view. Left ventricular mass (LVM) was calculated according to the Devereux formula: $LVM (g) = 0.8(1.04[(LVEDD + IVSd + PWd)^3 - LVEDD^3]) + 0.6$.¹² Left ventricular volumes were calculated using the biplane Simpson method of discs, by manual tracing (papillary muscles included in the cavity) in two-dimensional end-diastolic and end-systolic frames defined as the largest and smallest left ventricular cavities, respectively, in apical four-chamber and two-chamber projections. Ejection fraction (EF) was calculated automatically from end-diastolic volume (EDV) and end-systolic volume (ESV) using the following formula: $EF = (EDV - ESV) / EDV$. For assessment of left atrium (LA) volumes, the biplane area-length method was used: $LA \text{ volume} = (0.85 \times LA \text{ Area } 4 \text{ ch} \times LA \text{ Area } 2 \text{ ch}) / (\text{Longest atrial length})$. The values were indexed to body surface area. The LA endocardial borders were manually traced in both apical four-chamber and two-chamber views. Right atrium volumes were obtained using a single-plane disc summation technique in a dedicated apical four-chamber view.

Echocardiographic measurements were available in 198 of the study subjects with full data on age, sex, and biomarkers.

Endpoint assessment

Mortality was defined as all-cause mortality during the follow-up and was retrieved from the Swedish National Board of Health and Welfare's Cause of Death Register. Data regarding the re-hospitalization due to cardiac causes were retrieved from the individual electronic medical records of the Skåne Health Care Region (Medior, Siemens Health Services, Solna, Sweden), which cover all the citizens in the study catchment area.

Statistics

The variables are presented as means (\pm standard deviation) or median [25th–75th inter-quartile range (IQR)]. All variables that were not normally distributed were log transformed (NT-proBNP, cystatin C, copeptin, MR-proADM, and CT-pro-ET-1). Multivariable-adjusted Cox regression models were applied and log transformed, and standardized values of NT-proBNP, cystatin C, copeptin, MR-proADM, and CT-pro-ET-1 were entered as independent variables. Model 1 included age and sex, whereas Model 2 included age, sex, body mass index, diabetes status, smoking, presence of atrial fibrillation, systolic

blood pressure at admission, total cholesterol, high-density lipoprotein, and New York Heart Association class at admission. The time variable was calculated as follow-up time between screening and date of the first re-hospitalization, death, or end of follow-up through 1 October 2017. All analyses were performed using SPSS Windows Version 23.0, and a two-sided Bonferroni-corrected P -value of $0.05/5 = 0.010$ was considered statistically significant in the Cox regression analysis.

Echocardiographic measurements of cardiac dimensions and hypertrophy (eight different modalities) were tested for possible associations with the five biomarkers in age-adjusted and sex-adjusted linear regression analysis, and a two-sided Bonferroni-corrected P -value of $0.05/13 = 0.0038$ was considered statistically significant.

Results

The study population had a mean age of 75 years, were predominantly male (71%), 39% had diabetes, and 59% had previous or prevalent atrial fibrillation at inclusion. A high percentage of the patients received treatment with beta-blockers (92%) and angiotensin-converting enzyme inhibitors/angiotensin II receptor antagonists (78%) (Table 1).

During follow-up period (median time, 17 months; IQR [8–29]), a total of 57 patients died. The most frequent cause of death was HF ($n = 21$) followed by cardiac arrest ($n = 7$), cancer ($n = 2$), and stroke ($n = 2$). The remaining death causes ($n = 21$) consisted of different diagnoses and were defined as 'other' in the database.

A total of 90 patients were re-hospitalized (median follow-up time, 5 months; IQR [1–12]) because of cardiac causes. The most common cardiac causes of re-hospitalization were HF ($n = 79$) followed by cardiac arrhythmia ($n = 10$) and myocardial infarction ($n = 1$).

Biomarkers and mortality

In the Cox regression analyses adjusted for age and sex, all five biomarkers were significantly associated with increased post-discharge mortality (Table 2). In the multivariable analyses, all biomarkers, except CT-pro-ET-1, were significantly associated with mortality: cystatin C [hazard ratio (HR) 2.11, 95% confidence interval (CI) 1.56–2.86; $P = 1.0 \times 10^{-6}$], NT-proBNP (HR 1.85, 95% CI 1.17–2.17; $P = 4.0 \times 10^{-4}$), copeptin (HR 1.70, 95% CI 1.22–2.36; $P = 0.002$), and MR-proADM (HR 1.94, 95% CI 1.36–2.75; $P = 2.2 \times 10^{-4}$) (Table 2). Receiver operating characteristic curve analyses revealed Bonferroni-corrected significant associations for all biomarkers and all-cause mortality, except for CT-pro-ET-1: NT-proBNP (HR 0.669, 95% CI 0.590–0.749; $P < 0.001$), cystatin C (HR 0.722, 95% CI 0.649–0.794; $P < 0.001$), copeptin (HR 0.671,

Table 1 Baseline characteristics of the study population

	<i>n</i> = 268
Age (years)	75.1 (±11.0)
Sex (female), <i>n</i> (%)	77 (29)
Smoking, <i>n</i> (%)	31 (11.6)
BMI (kg/m ²)	27.4 (±5.6)
SBP (mmHg)	137.4 (±27.7)
DBP (mmHg)	79.2 (±15.3)
HT, <i>n</i> (%)	106 (39.6)
AHT, <i>n</i> (%)	268 (100%)
Beta-blockers, <i>n</i> (%)	137 (92)
ACEi or ARB, <i>n</i> (%)	208 (78)
Aldosterone antagonists, <i>n</i> (%)	14 (5)
Loop diuretics, <i>n</i> (%)	258 (96)
Diabetes, <i>n</i> (%)	105 (39)
Cholesterol (mmol/L)	3.6 (1.1)
HDL (mmol/L)	1.2 (0.4)
GFR (mL/min)	45.9 (16.8)
AF, <i>n</i> (%)	157 (58.6)
Newly diagnosed HF, <i>n</i> (%)	85 (32)
NT-proBNP (pmol/L)	4077.5 [2175.0–8125.8]
Cystatin C	1.6 [1.3–2.1]
Copeptin	30.9 [14.7–49.2]
MR-proADM	1.6 [1.1–2.2]
CT-pro-ET-1	149.3 [118.9–200.0]
LVEF, <i>n</i> (%)	39.1 (16.2)

ACEi, angiotensin-converting enzyme inhibitor; AF, atrial fibrillation; AHT, antihypertensive treatment; ARB, angiotensin II receptor antagonist; BMI, body mass index; CT-pro-ET-1, C-terminal pro-endothelin-1; DBP, diastolic blood pressure; GFR, glomerular filtration rate; HDL, high-density lipoprotein; HF, heart failure; HT, hypertension; LVEF, left ventricular ejection fraction; MR-proADM, mid-regional pro-adrenomedullin; NT-proBNP, N-terminal pro-brain natriuretic peptide. Values are means (± standard deviation) or median [25th–75th inter-quartile range].

95% CI 0.597–0.745; $P < 0.001$), MR-proADM (HR 0.655, 95% CI 0.575–0.734; $P < 0.001$), and CT-pro-ET-1 (HR 0.589, 95% CI 0.506–0.673; $P = 0.036$).

Biomarkers and re-hospitalization

In the age-adjusted and sex-adjusted Cox regression analysis, cystatin C and NT-proBNP were the only two of the five biomarkers significantly associated with risk of re-hospitalizations due to cardiac causes (Table 3). In the fully adjusted Cox regression Model 2, NT-proBNP was the only biomarker that showed significant association with risk of re-hospitalization due to cardiac causes (HR 1.43, 95% CI 1.10–1.87; $P = 0.009$) (Table 3). Receiver operating characteristic curve analyses revealed Bonferroni-corrected significant area under the curve (AUC) associations for all biomarkers and re-hospitalization, except for borderline significant MR-proADM: NT-proBNP (AUC 0.595, 95% CI 0.525–0.666; $P = 0.010$), cystatin C (AUC 0.614, 95% CI 0.542–0.687; $P = 0.002$), copeptin (AUC 0.599, 95% CI 0.530–0.668; $P = 0.007$), MR-proADM (AUC 0.597, 95% CI 0.524–0.669; $P = 0.011$), and CT-pro-ET-1 (AUC 0.606, 95% CI 0.537–0.675; $P = 0.004$).

Table 2 Cardiac biomarkers and risk of all-cause mortality

	Total mortality	
	HR (95% CI)	<i>P</i> -value
Cystatin C (<i>n</i> = 258; 56 events)		
Model 1	1.99 (1.52–2.62)	5.8×10^{-7}
Model 2	2.11 (1.56–2.86)	1.0×10^{-6}
NT-proBNP (<i>n</i> = 262; 55 events)		
Model 1	1.88 (1.37–2.57)	8.2×10^{-5}
Model 2	1.85 (1.32–2.61)	4.0×10^{-4}
Copeptin (<i>n</i> = 259; 57 events)		
Model 1	1.63 (1.20–2.20)	0.002
Model 2	1.70 (1.22–2.36)	0.002
MR-proADM (<i>n</i> = 250; 53 events)		
Model 1	1.78 (1.32–2.41)	1.9×10^{-4}
Model 2	1.94 (1.36–2.75)	2.2×10^{-4}
CT-pro-ET-1 (<i>n</i> = 260; 57 events)		
Model 1	1.45 (1.08–1.95)	0.014
Model 2	1.42 (1.03–1.95)	0.034

CI, confidence interval; CT-pro-ET-1, C-terminal pro-endothelin-1; HR, hazard ratio; MR-proADM, mid-regional pro-adrenomedullin; NT-proBNP, N-terminal pro-brain natriuretic peptide.

Cox regressions: Model 1: adjusted for age and sex; Model 2: adjusted for age, sex, body mass index, diabetes status, smoking, atrial fibrillation, systolic blood pressure at admission, total cholesterol, high-density lipoprotein, and New York Heart Association class at admission.

Biomarkers and echocardiographic measurements

High plasma NT-proBNP levels were robustly associated with reduced systolic left ventricular function as measured by EF (Table 4). MR-proADM and CT-pro-ET-1 were significantly associated with increased right ventricular size (Table 4). Finally, high levels of cystatin C were significantly associated with left posterior left ventricular wall hypertrophy and borderline associated with interventricular septum hypertrophy (Table 4).

Discussion

In this study, among patients hospitalized for newly diagnosed or exacerbated HF, elevated plasma levels of NT-proBNP, cystatin C, copeptin, and MR-proADM were significantly associated with increased post-discharge mortality on top of traditional cardiovascular risk factors. The only biomarker significantly associated with risk of re-hospitalization was NT-proBNP.

Cystatin C

Cystatin C is an established sensitive marker of glomerular filtration¹³ and a well-known predictor of cardiovascular disease.¹⁴ In HF patients, the predictive value of cystatin C in regard to mortality is higher compared with creatinine.¹⁵

Table 3 Cardiac biomarkers and risk of re-hospitalization

	1st re-hospitalization	
	HR (95% CI)	P-value
Cystatin C (n = 257; 89 events)		
Model 1	1.33 (1.08–1.65)	0.008
Model 2	1.27 (1.01–1.59)	0.040
NT-proBNP (n = 261; 89 events)		
Model 1	1.39 (1.10–1.77)	0.007
Model 2	1.43 (1.10–1.87)	0.009
Copeptin (n = 258; 88 events)		
Model 1	1.20 (0.96–1.49)	0.115
Model 2	1.20 (0.94–1.53)	0.152
MR-proADM (n = 249; 81 events)		
Model 1	1.28 (1.02–2.61)	0.031
Model 2	1.22 (0.93–1.596)	0.150
CT-pro-ET-1 (n = 260; 89 events)		
Model 1	1.30 (1.04–1.62)	0.019
Model 2	1.22 (0.95–1.57)	0.115

CI, confidence interval; CT-pro-ET-1, C-terminal pro-endothelin-1; HR, hazard ratio; MR-proADM, mid-regional pro-adrenomedullin; NT-proBNP, N-terminal pro-brain natriuretic peptide.

Cox regressions: Model 1: adjusted for age and sex; Model 2: adjusted for age, sex, body mass index, diabetes status, smoking, atrial fibrillation, systolic blood pressure at admission, total cholesterol, high-density lipoprotein, and New York Heart Association class at admission.

However, although these studies indicate that cystatin C might serve as a marker of disease susceptibility, a causal involvement has been repeatedly counter-proven in Mendelian randomization analyses.^{16,17} Hence, high levels of cystatin C are only regarded as robust markers of kidney function. In this context, it is not unexpected to see that higher levels of cystatin C are strongly associated with mortality in our study. Poor renal function has been shown to increase risk of cardiac remodelling (e.g. left ventricular hypertrophy),¹⁸ and indeed, we found an association between cystatin C and left ventricular hypertrophy, which in itself is a strong predictor of mortality.¹⁹ Moreover, the median glomerular filtration rate value in our cohort was low (mean 45 mL/min), and left ventricular EF was 39%, further implying an interplay between cardiac and renal dysfunction, often referred to as the cardio-renal syndrome.

N-terminal pro-brain natriuretic peptide

N-terminal pro-brain natriuretic peptide is an established biomarker reflecting HF severity and has earlier been associated with adverse outcomes in various HF populations.²⁰ In our study, NT-proBNP was the only biomarker that predicted re-hospitalization. Our results are in line with previous reports where elevated mature BNP levels predicted 30 day readmission for HF in over 50 000 subjects.²¹ Our finding that NT-proBNP is associated with reduced left ventricular function is concordant with previous studies.²²

Table 4 Biomarkers associated with echocardiographic measurements

	Cystatin C (n = 189)		NT-proBNP (n = 191)		Copeptin (n = 192)		MR-proADM (n = 185)		CT-pro-ET-1 (n = 192)	
	β (SE)	P-value	β (SE)	P-value	β (SE)	P-value	β (SE)	P-value	β (SE)	P-value
EF (%)	0.54 (1.26)	0.67	-7.07 (1.09)	8.6×10^{-10}	-2.22 (1.23)	0.07	-0.99 (1.23)	0.42	-1.05 (1.11)	0.35
IVSDd (mm/mm ²)	0.74 (0.26)	0.004	0.26 (0.25)	0.29	0.31 (0.26)	0.25	0.33 (0.25)	0.20	0.25 (0.24)	0.30
LVIDd (mm/m ²)	-2.02 (0.79)	0.01	-0.67 (0.77)	0.39	-1.04 (0.80)	0.20	-2.00 (0.77)	0.01	-1.48 (0.72)	0.04
RVIDd (mm/m ²)	0.24 (0.54)	0.66	-0.40 (0.51)	0.42	0.58 (0.53)	0.28	1.67 (0.50)	0.001	1.45 (0.47)	0.002
PWDD (mm/m ²)	1.81 (0.55)	0.001	0.90 (0.53)	0.09	1.17 (0.55)	0.04	1.61 (0.55)	0.004	1.33 (0.50)	0.008
LA volume (mL/m ²)	-1.69 (1.78)	0.34	-0.35 (1.67)	0.84	-0.36 (1.75)	0.84	-1.07 (1.74)	0.54	0.61 (1.58)	0.70
RA volume (mL/m ²)	-3.23 (2.08)	0.12	4.05 (1.93)	0.04	2.06 (2.06)	0.32	4.90 (2.00)	0.015	4.23 (1.84)	0.02
LVMl (g/m ²)	1.75 (3.24)	0.56	1.94 (2.97)	0.52	-1.23 (3.20)	0.70	-4.21 (3.08)	0.17	-3.69 (2.87)	0.20

CT-pro-ET-1, C-terminal pro-endothelin-1; EF, ejection fraction; IVSDd, interventricular systolic diameter diastole; LA, left atrium; LVIDd, left ventricular inner diameter diastole; LVMl, left ventricular mass index; MR-proADM, mid-regional pro-adrenomedullin; NT-proBNP, N-terminal pro-brain natriuretic peptide; PWDD, posterior wall diameter diastole; RA, right atrium; RVIDd, right ventricular inner diameter diastole.

β are unstandardized coefficients. Linear regressions are adjusted for age and sex.

Copeptin

Copeptin is located in the C-terminal section of the arginine vasopressin precursor and is a long-term stable pro-arginine vasopressin surrogate marker.²³ It has been implicated in poor outcome and mortality in numerous diseases such as diabetes,²⁴ myocardial infarction,²⁵ and stroke.²⁶ Copeptin is highly prognostic of 90 day adverse events in patients with acute HF, adding prognostic value to clinical predictors.²⁷ A recent meta-analysis comprising 10 prospective cohort studies demonstrated that the predictive value of copeptin is comparable with NT-proBNP for all-cause mortality in HF patients.²⁸ However, the possible use of copeptin as a target in biomarker-guided therapy in clinical practice remains to be investigated.

Mid-regional pro-adrenomedullin

Adrenomedullin is a hormone with vasodilatory, natriuretic, and hypotensive effects.²⁹ As adrenomedullin is an unstable hormone, its mid-regional prohormone fragment (MR-proADM) is more suitable for measurements, and its concentrations are correlated with those of adrenomedullin. A recent trial that included HF patients demonstrated that MR-proADM predicted mortality within 2 weeks superiorly to both mature BNP and NT-proBNP. When copeptin and MR-proADM were combined, the 14 day mortality prediction improved additionally. In a model where MR-proADM was added to BNP/NT-proBNP, the prediction of 90 day mortality significantly improved.³⁰ The association between MR-proADM and right ventricular size deserves further comment. Adrenomedullin has previously been proposed to participate in the mechanism that counteracts hypertension in the pulmonary circulation.³¹ Compensatory elevated levels of MR-proADM can therefore be expected in conditions that are associated with elevated pulmonary arterial pressure (e.g. decompensated HF) and consequently, right ventricular dilation, as seen here.

C-terminal pro-endothelin-1

Endothelin 1 is a potent vasopressor peptide and positive inotrope that has been implicated in myocardial infarction,³² hypertension,³³ and HF.³⁴ The C-terminal fragment of the endothelin-1 prohormone peptide (CT-pro-ET-1) is a stable surrogate marker for the instable ET-1. A recent meta-analysis focused on ET-1, pro-endothelin-1, and CT-pro-ET-1 demonstrated that increased levels of all three isoforms of the endothelin family were associated with poor prognosis or mortality in HF populations.³⁵

In the light of increased ET-1 levels and their association with elevated pulmonary vascular resistance, which in turn

leads to right ventricular dilatation and failure, our association between CT-pro-ET-1 levels and right ventricular diameter are sound and logical. As ET-1 is highly expressed in the lung, it can contribute to a strong vasoconstriction of the pulmonary arteries and veins and, consequently, to pulmonary hypertension.

In summary, progression of HF evokes abnormal neurohormonal compensatory responses. Measurements of biomarkers of neurohormonal systems could serve as novel tools for risk prediction in HF patients. However, it is necessary to emphasize that their clinical utility should be a target of further exploration. Taken together, our findings consolidate some of the prior observations on cardiovascular risk biomarkers and their predictive potential in relation to adverse outcomes in HF patients.

Study limitations

There are several strengths and limitations to this study. As we included patients admitted for new or worsening HF, with inability to deliver informed consent to the study as only exclusion criteria, our study population is most likely representative of the real-life clinical experience. However, our data were collected at a single centre, and the sample size was relatively small, which limits their applicability to other populations of HF patients. Moreover, the subjects included in HARVEST—Malmö were mainly of European descent, and the conclusions drawn might not be generalizable to all ancestries.

Conclusions

Among patients hospitalized for HF, elevated plasma levels of NT-proBNP, MR-proADM, copeptin, and cystatin C are significantly associated with increased post-discharge mortality, whereas NT-proBNP is the only biomarker that independently predicts the risk of re-hospitalization.

Acknowledgements

We thank the research nurses Hjördis Jernhed and Dina Chatziapostolou for their valuable contributions, and we thank all the staff at the echocardiographic laboratory at Skåne University Hospital, Malmö. The Knut och Alice Wallenbergs Stiftelse is acknowledged for generous support.

Conflict of interest

None declared.

Ethics statement

The study was approved by the ethics committee at Lund University, Sweden. All participants in this study signed a written informed consent form.

Data availability statement

Data will be available upon request.

Funding

Dr Magnusson was supported by grants from the Medicinska Fakulteten, Lunds Universitet, Skåne University Hospital, the Crafoord Foundation, the Ernhold Lundström Stiftelse, the Region Skåne, the Hulda och E Conrad Mossfelts Stiftelse för Vetenskaplig Forskning Inom Hjärt- och Kärlsjukdomarnas Område, the Southwest Skåne Diabetes Foundation, the Kockska Foundation, the Research Funds of Region Skåne, the Hjärt-Lungfonden, and the Wallenberg Center for Molecular Medicine, Lund University.

References

- Blecker S, Paul M, Taksler G, Ogedegbe G, Katz S. Heart failure-associated hospitalizations in the United States. *J Am Coll Cardiol* 2013; **61**: 1259–1267.
- Melander O, Newton-Cheh C, Almgren P, Hedblad B, Berglund G, Engstrom G, Persson M, Smith JG, Magnusson M, Christensson A, Struck J, Morgenthaler NG, Bergmann A, Pencina MJ, Wang TJ. Novel and conventional biomarkers for prediction of incident cardiovascular events in the community. *JAMA* 2009; **302**: 49–57.
- Demissei BG, Postmus D, Cleland JG, O'Connor CM, Metra M, Ponikowski P, Teerlink JR, Cotter G, Davison BA, Givertz MM, Bloomfield DM, van Veldhuisen DJ, Dittrich HC, Hillege HL, Voors AA. Plasma biomarkers to predict or rule out early post-discharge events after hospitalization for acute heart failure. *Eur J Heart Fail* 2017; **19**: 728–738.
- Christensson A, Grubb A, Molvin J, Holm H, Gransbo K, Tasevska-Dinevska G, Bachus E, Jujic A, Magnusson M. The shrunken pore syndrome is associated with declined right ventricular systolic function in a heart failure population—the HARVEST study. *Scand J Clin Lab Invest* 2016; **76**: 568–574.
- Ali A, Holm H, Molvin J, Bachus E, Tasevska-Dinevska G, Fedorowski A, Jujic A, Magnusson M. Autonomic dysfunction is associated with cardiac remodelling in heart failure patients. *ESC Heart Fail* 2017; **5**: 46–52.
- Williams B, Mancia G, Spiering W, Agabiti Rosei E, Azizi M, Burnier M, Clement DL, Coca A, de Simone G, Dominiczak A, Kahan T, Mahfoud F, Redon J, Ruilope L, Zanchetti A, Kerins M, Kjeldsen SE, Kreutz R, Laurent S, Lip GYH, McManus R, Narkiewicz K, Ruschitzka F, Schmieder RE, Shlyakhto E, Tsioufis C, Aboyans V, Desormais I, ESC Scientific Document Group. 2018 ESC/ESH guidelines for the management of arterial hypertension. *Eur Heart J* 2018; **39**: 3021–3104.
- Fenske W, Stork S, Blechschmidt A, Maier SG, Morgenthaler NG, Allolio B. Copeptin in the differential diagnosis of hyponatremia. *J Clin Endocrinol Metab* 2009; **94**: 123–129.
- Morgenthaler NG, Struck J, Alonso C, Bergmann A. Measurement of midregional proadrenomedullin in plasma with an immunoluminometric assay. *Clin Chem* 2005; **51**: 1823–1829.
- Caruhel P, Mazier C, Kunde J, Morgenthaler NG, Darbouret B. Homogeneous time-resolved fluoroimmunoassay for the measurement of midregional proadrenomedullin in plasma on the fully automated system B.R.A.H.M.S KRYPTOR. *Clin Biochem* 2009; **42**: 725–728.
- Papassotiropoulos J, Morgenthaler NG, Struck J, Alonso C, Bergmann A. Immunoluminometric assay for measurement of the C-terminal endothelin-1 precursor fragment in human plasma. *Clin Chem* 2006; **52**: 1144–1151.
- Recommendations for cardiac chamber quantification by echocardiography in adults: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *Eur Heart J Cardiovasc Imaging* 2016; **17**: 412.
- Devereux RB, Alonso DR, Lutas EM, Gottlieb GJ, Campo E, Sachs I, Reichek N. Echocardiographic assessment of left ventricular hypertrophy: comparison to necropsy findings. *Am J Cardiol* 1986; **57**: 450–458.
- Dharmidharka VR, Kwon C, Stevens G. Serum cystatin C is superior to serum creatinine as a marker of kidney function: a meta-analysis. *Am J Kidney Dis* 2002; **40**: 221–226.
- Shlipak MG, Sarnak MJ, Katz R, Fried LF, Seliger SL, Newman AB, Siscovick DS, Stehman-Breen C. Cystatin C and the risk of death and cardiovascular events among elderly persons. *N Engl J Med* 2005; **352**: 2049–2060.
- Shlipak MG, Katz R, Fried LF, Jenny NS, Stehman-Breen CO, Newman AB, Siscovick D, Psaty BM, Sarnak MJ. Cystatin-C and mortality in elderly persons with heart failure. *J Am Coll Cardiol* 2005; **45**: 268–271.
- Svensson-Farbom P, Almgren P, Hedblad B, Engstrom G, Persson M, Christensson A, Melander O. Cystatin C is not causally related to coronary artery disease. *PLoS One* 2015; **10**: e0129269.
- O'Seaghdha CM, Tin A, Yang Q, Katz R, Liu Y, Harris T, Astor B, Coresh J, Fox CS, Kao WH, Shlipak MG. Association of a cystatin C gene variant with cystatin C levels, CKD, and risk of incident cardiovascular disease and mortality. *Am J Kidney Dis* 2014; **63**: 16–22.
- Di Lullo L, Gorini A, Russo D, Santoboni A, Ronco C. Left ventricular hypertrophy in chronic kidney disease patients: from pathophysiology to treatment. *Cardiorenal Med* 2015; **5**: 254–266.
- Vakili BA, Okin PM, Devereux RB. Prognostic implications of left ventricular hypertrophy. *Am Heart J* 2001; **141**: 334–341.
- Doust JA, Pietrzak E, Dobson A, Glasziou P. How well does B-type natriuretic peptide predict death and cardiac events in patients with heart failure: systematic review. *BMJ* 2005; **330**: 625.
- Flint KM, Allen LA, Pham M, Heidenreich PA. B-type natriuretic peptide predicts 30-day readmission for heart failure but not readmission for other causes. *J Am Heart Assoc* 2014; **3**: e000806.
- Krishnaswamy P, Lubien E, Clopton P, Koon J, Kazanegra R, Wanner E, Gardetto N, Garcia A, DeMaria A, Maisel AS. Utility of B-natriuretic peptide levels in identifying patients with left ventricular systolic or diastolic dysfunction. *Am J Med* 2001; **111**: 274–279.
- Holwerda DA. A glycopeptide from the posterior lobe of pig pituitaries. I. Isolation and characterization. *Eur J Biochem* 1972; **28**: 334–339.
- Velho G, El Boustany R, Lefèvre G, Mohammedi K, Fumeron F, Potier L, Bankir L, Bouby N, Hadjadj S, Marre

- M, Roussel R. Plasma copeptin, kidney outcomes, ischemic heart disease, and all-cause mortality in people with long-standing type 1 diabetes. *Diabetes Care* 2016; **39**: 2288–2295.
25. Raskovalova T, Twerenbold R, Collinson PO, Keller T, Bouvaist H, Folli C, Giavarina D, Lotze U, Eggers KM, Dupuy AM, Chenevier-Gobeaux C, Meune C, Maisel A, Mueller C, Labarère J. Diagnostic accuracy of combined cardiac troponin and copeptin assessment for early rule-out of myocardial infarction: a systematic review and meta-analysis. *Eur Heart J Acute Cardiovasc Care* 2014; **3**: 18–27.
26. Choi KS, Kim HJ, Chun HJ, Kim JM, Yi HJ, Cheong JH, Kim CH, Oh SJ, Ko Y, Kim YS, Bak KH, Ryu JI, Kim W, Lim T, Ahn H, Ahn IM, Lee SH Prognostic role of copeptin after stroke: a systematic review and meta-analysis of observational studies. *Sci Rep* 2015; **5**: 11665.
27. Maisel A, Xue Y, Shah K, Mueller C, Nowak R, Peacock WF, Ponikowski P, Mockel M, Hogan C, Wu AH, Richards M, Clopton P, Filippatos GS, Di Somma S, Anand IS, Ng L, Daniels LB, Neath SX, Christenson R, Potocki M, McCord J, Terracciano G, Kremastinos D, Hartmann O, von Haehling S, Bergmann A, Morgenthaler NG, Anker SD. Increased 90-day mortality in patients with acute heart failure with elevated copeptin: secondary results from the Biomarkers in Acute Heart Failure (BACH) study. *Circ Heart Fail* 2011; **4**: 613–620.
28. Zhong Y, Wang R, Yan L, Lin M, Liu X, You T. Copeptin in heart failure: review and meta-analysis. *Clin Chim Acta* 2017; **475**: 36–43.
29. Jougasaki M, Burnett JC Jr. Adrenomedullin: potential in physiology and pathophysiology. *Life Sci* 2000; **66**: 855–872.
30. Maisel A, Mueller C, Nowak R, Peacock WF, Landsberg JW, Ponikowski P, Mockel M, Hogan C, Wu AH, Richards M, Clopton P, Filippatos GS, Di Somma S, Anand I, Ng L, Daniels LB, Neath SX, Christenson R, Potocki M, McCord J, Terracciano G, Kremastinos D, Hartmann O, von Haehling S, Bergmann A, Morgenthaler NG, Anker SD. Mid-region pro-hormone markers for diagnosis and prognosis in acute dyspnea. *J Am Coll Cardiol* 2010; **55**: 2062.
31. Yoshihara F, Nishikimi T, Horio T, Yutani C, Takishita S, Matsuo H, Ohe T, Kangawa K. Chronic infusion of adrenomedullin reduces pulmonary hypertension and lessens right ventricular hypertrophy in rats administered monocrotaline. *Eur J Pharmacol* 1998; **355**: 33–39.
32. Tomoda H. Plasma endothelin-1 in acute myocardial infarction with heart failure. *Am Heart J* 1993; **125**: 667–672.
33. Schiffrin EL. Role of endothelin-1 in hypertension and vascular disease. *Am J Hypertens* 2001; **14**: 83S–89S.
34. Wei CM, Lerman A, Rodeheffer RJ, McGregor CG, Brandt RR, Wright S, Heublein DM, Kao PC, Edwards WD, Burnett JC Jr. Endothelin in human congestive heart failure. *Circulation* 1994; **89**: 1580–1586.
35. Zhang C-L, Xie S, Qiao X, An Y-M, Zhang Y, Li L, Guo XB, Zhang FC, Wu LL. Plasma endothelin-1-related peptides as the prognostic biomarkers for heart failure: a PRISMA-compliant meta-analysis. *Medicine* 2017; **96**: e9342-e.