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Prognostic Value of Serial ST2 Measurements in Patients With Acute Heart Failure

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ABSTRACT

BACKGROUND Several clinical studies have evaluated the association between ST2 and outcome in patients with heart failure (HF). However, little is known about the predictive value of frequently measured ST2 levels in patients with acute HF.

OBJECTIVES This study sought to describe the prognostic value of baseline and repeated ST2 measurements in patients with acute HF.

METHODS In the TRIUMPH (Translational Initiative on Unique and novel strategies for Management of Patients with Heart failure) clinical cohort study, 496 patients with acute HF were enrolled in 14 hospitals in the Netherlands between 2009 and 2014. Repeated blood samples (7) were drawn during 1-year follow-up. ST2 and N-terminal pro–B-type natriuretic peptide (NT-proBNP) levels were measured in a central laboratory. The primary endpoint was the composite of all-cause mortality and HF rehospitalization. Associations between repeated biomarker measurements and the primary endpoint were assessed using a joint model.

RESULTS Median age was 74 years, and 37% of patients were women. The primary endpoint was reached in 188 patients (40%) during a median follow-up of 325 days (interquartile range: 85 to 401). The median baseline ST2 level was 71 ng/ml (interquartile range: 46 to 102). After adjustment for clinical factors and NT-proBNP, baseline ST2 was associated with an increased risk of the primary endpoint, and the hazard ratio per 1 SD increase of the baseline ST2 level (on the $\log_2$ scale) was 1.30 (95% confidence interval: 1.08 to 1.56; $p = 0.005$). When repeated measurements were taken into account, the adjusted hazard ratio per 1 SD increase of the ST2 level (on the $\log_2$ scale) during follow-up increased to 1.85 (95% confidence interval: 1.02 to 3.33; $p = 0.044$), adjusted for clinical factors and repeated measurements of NT-proBNP. Furthermore, ST2 levels appeared to elevate several weeks before the time of the primary endpoint.

CONCLUSIONS Repeated ST2 measurements appeared to be a strong predictor of outcome in patients with acute HF, independent of repeatedly measured NT-proBNP. Hence ST2 may be helpful in clinical practice for prognostication and treatment monitoring. (TRanslational Initiative on Unique and novel strategies for Management of Patients with Heart failure [TRIUMPH]; NTR1893) (J Am Coll Cardiol 2017;70:2378–88) © 2017 by the American College of Cardiology Foundation.

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Heart failure (HF) is a major cause of morbidity and mortality in the Western World (1). Improvements in treatment and patient management are needed because most patients with HF die despite evidence-based treatment. Serum biomarkers may play an important role in bridging the gap between the assessment of HF and the occurrence of adverse outcomes, and they may expose novel, potentially modifiable disease pathways.

Most studies on the prognostic value of biomarkers of HF conducted so far have related adverse outcome during follow-up with a single measurement at baseline (2–4). This approach does not explore the biological variation that exists within patients with a highly variable, heterogeneous, and progressive condition such as HF (5). Thus, repeated biomarker measurements may be required to reflect more accurately the dynamic and progressive nature of the underlying pathophysiological processes, such as mechanical overload, cardiac fibrosis, and inflammation, and therefore may be more suitable for prognostication and therapy monitoring.

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ST2 is an interleukin-1 (IL-1) receptor family member with membrane-bound (ST2L) and soluble (sST2) isoforms. An IL-1-related protein, called IL-33, was identified as a functional ligand for ST2L (6). IL-33/ST2L signaling protects the myocardium against hypertrophy and cardiac fibrosis following pressure overload (7). Soluble ST2, which is the form measured by current assays, acts as a decoy receptor for IL-33 and prevents the IL-33/ST2L interaction and the subsequent cardioprotective cascade of events. The major source of ST2 is currently not fully established. For a long time, the source of circulating sST2 in cardiac disease was presumed to be myocardial, following in vitro data that sST2 has been shown to be secreted by cardiomyocytes when the cells are subjected to biomechanical overload (8). Accordingly, serum ST2 levels correlate strongly with serum levels of natriuretic peptides (9). More recent work, however, suggests that in human cardiac disease, the vascular endothelial cells may be the predominant source of sST2, rather than the human myocardium (10).

In clinical studies, single ST2 levels have shown to be a risk factor for mortality in patients with both stable and acute HF, independent of N-terminal pro-B-type (NT-proBNP) (2,11,12). A recent meta-analysis supports the use of ST2 in patients with stable chronic HF for risk stratification (12). Furthermore, several studies have evaluated the prognostic value of multiple ST2 measurements (9,13–15). It is known that ST2 levels in patients with acute HF are significantly higher than in patients with chronic HF and fall rapidly over days to weeks during HF treatment (13). This lack of reduction in ST2 level during acute HF treatment is predictive of mortality. In addition, persistently high levels of ST2 were associated with increased mortality risk (16). Only a few studies, most in patients with chronic systolic HF, have evaluated the prognostic value of the change in ST2 levels, in which the ST2 level was measured with an interval of at least 1 month (14,15). Increases in ST2 levels from baseline to 12 months were associated with a significant increased risk for all-cause mortality. On the contrary, the CORONA study (Controlled Rosuvastatin Multinational Trial in Heart Failure) showed that change in ST2 levels from baseline to 3 months was not associated with mortality (17). The RELAX-AHF (Efficacy and Safety of Relaxin for the Treatment of Acute Heart Failure) trial showed that serial sST2 measurements combined in a multimarker approach are useful for prognostication in patients with acute HF (18).

Given the dynamic and progressive nature of HF and the pathophysiology of ST2, we hypothesized that in patients admitted with acute HF, frequently measured ST2 levels during follow-up will add incremental prognostic information to that conferred by repeated measurements of NT-proBNP. In the American Heart Association/American College of Cardiology guidelines for management of heart failure, ST2 is considered useful for prognostication and therapy monitoring, but more research is required to support this suggestion (19). Therefore, in the present TRIUMPH study (TRanslational Initiative on Unique and novel strategies for Management of Patients with Heart failure [TRIUMPH]: NTR1893), we assessed the association between frequently measured ST2 independent of frequently measured NT-proBNP and the incidence of all-cause mortality and HF readmission during 1-year follow-up in 496 patients admitted with acute HF.

ABBREVIATIONS AND ACRONYMS

ACC = American College of Cardiology

AHA = American Heart Association

CI = confidence interval

eGFR = estimated glomerular filtration rate

HF = heart failure

HR = hazard ratio

IL = interleukin

IQR = interquartile range

LVEF = left ventricular ejection fraction

NT-pro-BNP = N-terminal pro-B-type natriuretic peptide

METHODS

OBJECTIVE AND STUDY DESIGN. TRIUMPH was designed as a translational bench-to-bedside study program encompassing the entire spectrum of biomarker discovery to clinical validation. The clinical validation study was an observational prospective study enrolling patients admitted with acute HF in 14 hospitals in the Netherlands between September 2009 and December 2013. This cohort study was

designed to validate the clinical value of biomarkers successfully passing the bioinformatics and early validation stages of TRIUMPH, as well as to evaluate more established biomarkers of HF further. There was a particular interest in the change in biomarker levels over time, as well as in the analyses and prognostic significance of repeated biomarker sampling during the follow-up of patients with HF. The study was approved by the medical ethics committees at all participating centers.

PATIENT SELECTION. Patients ≥18 years of age were eligible for enrollment if they were hospitalized with decompensation of known chronic HF or newly diagnosed HF. Furthermore, 3 other criteria had to be met: 1) natriuretic peptide levels had to be elevated to ≥3 times the upper limit of normal; 2) there had to be evidence of sustained systolic or diastolic left ventricular dysfunction; and 3) patients had to be treated with intravenous diuretics. Patients with HF that was precipitated by a noncardiac condition, by severe valvular dysfunction without sustained left ventricular dysfunction, or by an acute ST-segment elevation myocardial infarction were excluded. Furthermore, patients scheduled for a coronary revascularization procedure, on a waiting list for heart transplantation, with severe renal failure for which dialysis was needed, or with a coexisting condition with a life expectancy <1 year could not participate. All study participants provided written informed consent.

PATIENT MANAGEMENT. Patient management was at the discretion of the treating physician and was provided in accordance with the guidelines of the European Society of Cardiology (20). Importantly, the biomarker data that were generated in the context of this observational study were not used for treatment decisions.

STUDY PROCEDURES. During hospitalization, blood samples were obtained at admission (day 1), once during days 2 to 4, and subsequently on the day of discharge. Afterward, repeated blood samples were also obtained at outpatient follow-up visits, which were planned at 2 to 4 weeks, 3 months, 6 months, and 9 to 12 months after discharge. The baseline blood sample was defined as the first sample obtained after inclusion, up to a maximum of 2 days after inclusion. At each visit, HF symptoms were assessed using the New York Heart Association functional classification. Medication use was determined at discharge by using 3 categories: 1) use of an angiotensin-converting enzyme inhibitor or an angiotensin II receptor antagonist, or both; 2) use of a beta-blocker; or 3) use of diuretics. Patients underwent physical examination, and weight, blood pressure, and heart rate were systematically measured.

BLOOD COLLECTION. Nonfasting blood samples were obtained by venipuncture and transported to the clinical chemistry laboratory of each participating hospital for further processing according to a standardized protocol. The collected material was centrifuged at 1,700 G/relative centrifugal force, and then heparin plasma and blood serum were separated. All blood aliquots were subsequently stored at a temperature of −80°C within 2 h after venipuncture.

ST2 MEASUREMENTS. Serum samples and heparin plasma samples were transported under controlled conditions to a central laboratory (Future Diagnostics Solutions B.V., Wijchen, the Netherlands) for batch analysis of ST2 and NT-proBNP levels. ST2 concentrations were determined in serum in single measurements by using a quantitative sandwich monoclonal enzyme-linked immunosorbent assay (Presage ST2 Assay, Critical Diagnostics, Inc., San Diego, California). In our hands the average coefficient of variation for interassay variation was 4.9%, in line with the average interassay coefficient of variation of 5.2% reported by the manufacturer. NT-proBNP concentrations were determined in heparin plasma by using the Elecsys NT-proBNP electrochemiluminescent sandwich immunoassay on a Cobas 8000 analyzer (Roche Diagnostics, Ltd., Rotkreuz, Switzerland). Analysts were blinded to patients' characteristics and endpoints.

ST2 PATTERN. Post hoc analyses were performed to identify ST2 patterns in patients with and without the primary endpoint. Two investigators, blinded to baseline patients' characteristics and clinical outcomes data, individually analyzed the ST2 pattern. ST2 patterns were classified as follows: 1) "U-shaped," if the ST2 level initially decreased and later increased; 2) "J-shaped," if the ST2 level initially decreased and did not increase later; 3) "not interpretable," if fewer than 3 ST2 measurements were available or 3 ST2 measurements were close together; or 4) "other," if a different ST2 pattern was identified. If there was disagreement, a consensus was reached in a separate session.

ENDPOINTS. Information on vital status and hospital readmissions was obtained until at least 9 months with a maximum of 400 days after the index hospitalization. We approached the civil registry, screened all medical records, and asked patients for information during their follow-up visits.

The primary endpoint is the composite of all-cause mortality and readmission for HF. Readmission for

TABLE 1 Baseline Characteristics According to the Overall Sample (n = 475) and Quartiles of Baseline ST2 Level (n = 386)

	Overall Sample	Q 1	Q 2	Q 3	Q 4	p Value*
Demographic characteristics						
Age, yrs	74 (65-80)	72	75	73	74	0.427
Female	37	45	37	38	34	0.434
Caucasian	95	91	95	95	95	0.541
Measurements at baseline						
Body mass index, kg/m^2	28 (25-31)	28	28	28	27	0.768
Systolic blood pressure, mm Hg	125 (110-147)	128	135	124	124	0.534
Diastolic blood pressure, mm Hg	74 (65-85)	75	76	72	74	0.513
Heart rate, beats/min	85 (72-100)	85	86	84	84	0.503
eGFR, ml/min/1.73 m^2	46 (34-62)	51	49	44	40	0.002
Left ventricular ejection fraction, %	30 (21-41)	34	30	30	29	0.204
NYHA functional classification						0.378
II	17	20	16	16	11	
III	55	53	58	63	53	
IV	27	27	25	20	34	
Medical history						
Newly diagnosed heart failure	36	43	40	37	27	0.088
Heart failure with reduced ejection fraction	83	78	85	79	87	0.434
Previous heart failure admission within 6 months	20	20	18	15	27	0.245
Ischemic heart failure	49	43	44	47	53	0.498
Myocardial infarction	40	35	31	43	50	0.034
Hypertension	51	55	55	46	48	0.470
Atrial fibrillation	42	38	45	43	46	0.640
Diabetes mellitus	36	32	32	41	39	0.439
Stroke	17	13	16	16	19	0.718
Biomarkers						
ST2, ng/ml	71 (46-102)	37	59	89	132	
NT-proBNP, pg/ml	4,152 (2,089-9,387)	2,347	3,970	4,871	5,692	<0.001
Endpoints						
Primary endpoint	40	23	34	44	52	<0.001
All-cause mortality	24	7	20	26	32	<0.001
HF hospitalization	26	20	27	33	34	0.15
Cardiovascular mortality	16	2	15	17	23	<0.001

Values are median (interquartile range) or %. *p value for differences between quartiles of baseline ST2 level.
eGFR = estimated glomerular filtration rate; HF = heart failure; NT-pro-BNP = N-terminal pro-B-type natriuretic peptide; NYHA = New York Heart Association; Q = quartile.

HF was defined as an unplanned rehospitalization resulting from decompensation of HF, with at least 2 of the following 3 criteria being present: elevated natriuretic peptide levels ≥3 times the upper limit of normal; symptoms of cardiac decompensation (rales, edema, or elevated central venous pressure); and treatment with intravenous diuretics. Secondary endpoints included the individual components of the primary endpoint and cardiovascular mortality. An event adjudication committee, blinded to biomarker information, was established for reviewing and adjudication of endpoints.

STATISTICAL ANALYSIS. The distributions of continuous variables were evaluated for normality by visual examination of the histogram and Kolmogorov-Smirnov tests. Variables with a normal distribution are presented as mean ± SD, whereas the median and interquartile range (IQR) are presented in case of non-normality. Categorical variables are presented as counts and percentages. ST2 and NT-proBNP levels had a non-normal distribution and were therefore log-transformed for further analyses.

Patients were classified according to the quartiles of the ST2 distribution, and differences in baseline characteristics between these quartiles were evaluated by chi-square tests (categorical variables), analysis of variance, or Kruskal-Wallis tests, as appropriate.

We applied Cox proportional hazards models to evaluate the association of baseline ST2 levels with the study endpoints. Subjects were censored at the time of occurrence of the endpoint under investigation, death, and at the scheduled end of follow-up.

TABLE 2 Hazard Ratios for Different Endpoints per 1 SD Increase of the Baseline ST2 Level (on the Log_2 Scale)

	N	Baseline Level*	
		HR (95% CI)	p Value
Primary endpoint			
Model 1†		1.49 (1.26–1.77)	<0.001
Model 2		1.48 (1.25–1.76)	<0.001
Model 3		1.30 (1.08–1.56)	0.005
Number of events/patients	188/475		
All-cause mortality			
Model 1		1.80 (1.41–2.29)	<0.001
Model 2		1.77 (1.39–2.27)	<0.001
Model 3		1.43 (1.11–1.86)	0.006
Number of events/patients	113/475		
HF hospitalization			
Model 1		1.33 (1.09–1.61)	0.005
Model 2		1.33 (1.09–1.61)	0.005
Model 3		1.16 (0.94–1.43)	0.159
Number of events/patients	123/475		
Cardiovascular mortality			
Model 1		2.01 (1.49–2.72)	<0.001
Model 2		1.98 (1.46–2.67)	<0.001
Model 3		1.63 (1.19–2.23)	0.002
Number of events/patients	77/475		

Mean ± 1 SD of the patient-specific geometric mean ST2 value at baseline (presented on the linear scale): 70.0 (40.7 ± 120.3). *Hazard ratios are related to a 1 SD increase of ST2 (on the log scale) at baseline. †Model 1 unadjusted; model 2 adjusted for age and sex; model 3 adjusted for age, sex, systolic blood pressure, diabetes mellitus, left ventricular ejection fraction, previous hospitalization for HF during the last 6 months, ischemic heart failure, body mass index, estimated glomerular filtration rate, and baseline NT-proBNP.

CI = confidence interval; HF = heart failure; HR = hazard ratio; other abbreviations as in Table 1.

No deviations of the proportional hazards assumption were found by inspecting log minus log plots of the survival functions. We performed univariate analyses to obtain the crude estimates of the effect of baseline ST2 level (model 1), analyses that were adjusted for age and sex only (model 2), and analyses that were additionally adjusted for systolic blood pressure, diabetes mellitus, left ventricular ejection fraction (LVEF), previous hospitalization for HF during the last 6 months, ischemic HF, body mass index, estimated glomerular filtration rate (eGFR), and baseline NT-proBNP level (model 3). The results are presented as adjusted hazard ratios (HRs) per 1 SD increase of the biomarker level (on the log_2 scale) with 95% confidence intervals (CIs). We calculated the eGFR using the Modification of Diet in Renal Disease equation (21).

Joint models were fitted to assess the association between estimated instantaneous biomarker levels during follow-up, calculated using the repeated time-dependent biomarker levels, and the specified study endpoints. A joint model combines a mixed-effects linear regression model for the serial measurements with a Cox proportional hazards model for the risk of the specified study endpoints (22). We used cubic splines, with knots set at 1 week and 1 month after initial hospitalization, for the mixed model. For the analyses with the repeated ST2 measurements, we performed univariate analyses (model 1). We combined repeated measurements of ST2 and NT-proBNP in 1 joint model to assess their independent prognostic value and adjusted for age and sex (model 2). We additionally adjusted for systolic blood pressure, diabetes mellitus, LVEF, previous hospitalization for HF during the last 6 months, ischemic HF, body mass index, eGFR, and use of medication at hospital discharge (angiotensin-converting enzyme inhibitor and/or angiotensin II receptor antagonist, beta-blocker, diuretics) (model 3). We also tested whether the slope of the ST2 trajectories itself, when added to model 3, was an independent predictor. Diagnostics and sensitivity analyses were performed to evaluate the joint models. The final results are presented as adjusted HRs per 1 SD increase of the biomarker level (on the log_2 scale) at any point in time with 95% CIs. Data on covariates were complete in 93% of patients, except for LVEF, which was complete in 78%. Single imputation was applied to account for missing values of covariates.

Statistical Package for Social Sciences, version 21.0 software (SPSS, IBM Corp., Armonk, New York) was used for descriptive data analysis. R statistical software (version 2.15.0, R Foundation, Vienna, Austria) was used for advanced statistical analyses of the longitudinal biomarker data and study endpoints (packages JMBayes and JM). All statistical tests were 2-tailed, and p values <0.05 were considered statistically significant.

RESULTS

PATIENTS. A total of 496 patients were enrolled in the TRIUMPH clinical cohort. Three patients withdrew their informed consent. Eighteen patients were withdrawn from statistical analyses because of inclusion violation. These patients had no evidence of sustained systolic or diastolic left ventricular dysfunction on echocardiography. Accordingly, 475 patients comprised the analysis set. Their median age was 74 years (IQR: 65 to 80 years), and 37% were women (Table 1). Median systolic blood pressure was 125 mm Hg (IQR: 110 to 147 mm Hg), and median LVEF was 30% (IQR: 21% to 42%). Most patients had HF with a reduced ejection fraction (83%). The median baseline ST2 level was 71 ng/ml (IQR: 46 to 102 ng/ml), and that of NT-proBNP was 4,152 pg/ml (IQR: 2,089 to 9,387 pg/ml). Additionally, Table 1 shows the baseline characteristics of patients in different

quartiles of ST2 level. Patients in quartiles with a higher ST2 level had worse kidney function, and more patients had a history of myocardial infarction.

BASELINE ST2 LEVELS AND THE INCIDENCE OF STUDY ENDPOINTS. During the median follow-up of 325 days (IQR: 85 to 401 days), 188 patients (40%) reached the primary endpoint of all-cause death (n = 113) or readmission for HF (n = 123). This corresponds with an incidence rate of 55.9 per 100 patient-years for the primary endpoint. Baseline ST2 levels were available in 386 patients. In the highest quartile of baseline ST2, 50 patients (52%) reached the primary endpoint compared with 22 patients (23%) in the lowest quartile of ST2. All-cause mortality was also higher in the highest ST2 quartile compared with the lowest ST2 quartile: 31 (32%) and 7 (7%), respectively. This was similar for cardiovascular mortality: 22 (23%) and 2 (2%), respectively (Table 1).

The baseline ST2 level was associated with an increased risk of all the predefined study endpoints (Table 2). With respect to the primary endpoint, all-cause mortality and cardiovascular mortality, these associations remained statistically significant after adjustment for all selected potential confounders, including baseline NT-proBNP level (model 3).

PROGNOSTIC VALUE OF REPEATED ST2 MEASUREMENTS. The average number of ST2 measurements per patient during follow-up was 3.9 and 4.1 for NT-pro-BNP. After adjustment for repeated measurements of NT-pro-BNP, age, and sex (model 2), the HR for the primary endpoint corresponding to a 1 SD increase of ST2 level (on the $\log_2$ scale) during follow-up was 3.54 (95% CI: 2.07 to 7.32; $p < 0.001$). After adjustment for the broader range of potential confounders including repeated measurements of NT-proBNP (model 3), the association remained statistically significant, with an HR corresponding to a 1 SD increase of ST2 level (on the $\log_2$ scale) during follow-up of 1.85 (95% CI: 1.02 to 3.33; p 0.044). The HR corresponding to a 1 SD increase of NT-proBNP level (on the $\log_2$ scale) during follow-up for the primary endpoint was 2.13 (95% CI: 1.35 to 3.88; $p < 0.001$) adjusted for model 3 and repeated measurements of ST2 (Table 3). The HRs for all-cause and cardiovascular mortality corresponding to a 1 SD increase of ST2 level (on the $\log_2$ scale) during follow-up after adjustment for all covariates and repeated measurements of NT-proBNP (model 3) were highly statistically significant: 4.36 (95% CI: 2.31 to 8.92; $p < 0.001$) and 3.98 (95% CI: 2.15 to 7.94; $p < 0.001$), respectively. The slope of the ST2 level trajectories itself was not an independent predictor of the primary endpoint.

TABLE 3 Hazard Ratios for Different Endpoints per 1 SD Increase of ST2 Level or NT-proBNP Level (on the $\log_2$ Scale) at Any Point in Time Using Repeated ST2 and Repeated NT-proBNP Measurements in a Joint Model

	Model*	Mean Value† Mean ± SD	Instantaneous Level‡ HR (95% CI)	Instantaneous Level‡ p Value
Primary endpoint				
ST2 (crude)	1	41.4 (24.2 ± 70.9)	2.78 (2.16–3.64)	<0.001
ST2	2	41.4 (24.2 ± 70.9)	3.54 (2.07–7.32)	<0.001
NT-proBNP	2	1,776 (517 ± 6,093)	1.67 (1.20–2.34)	0.002
ST2	3	41.4 (24.2 ± 70.9)	1.85 (1.02–3.33)	0.044
NT-proBNP	3	1,776 (517 ± 6,093)	2.13 (1.35–3.88)	<0.001
All-cause mortality				
ST2 (crude)	1	42.6 (24.8 ± 73.3)	4.45 (3.12–6.39)	<0.001
ST2	2	42.6 (24.8 ± 73.3)	4.19 (2.31–8.79)	<0.001
NT-proBNP	2	1,874 (545 ± 6,447)	1.85 (1.22–2.83)	0.002
ST2	3	42.6 (24.8 ± 73.3)	4.36 (2.31–8.92)	<0.001
NT-proBNP	3	1,874 (545 ± 6,447)	2.48 (1.35–6.10)	0.004
HF hospitalization				
ST2 (crude)	1	41.4 (24.2 ± 70.9)	2.24 (1.68–3.01)	<0.001
ST2	2	41.4 (24.2 ± 70.9)	1.80 (1.27–2.56)	<0.001
NT-proBNP	2	1,776 (517 ± 6,093)	1.62 (1.18–2.19)	<0.001
ST2	3	41.4 (24.2 ± 70.9)	1.10 (0.64–1.83)	0.690
NT-proBNP	3	1,776 (517 ± 6,093)	1.47 (0.92–2.45)	0.096
Cardiovascular mortality				
ST2 (crude)	1	42.6 (24.8 ± 73.3)	5.27 (3.31–8.31)	<0.001
ST2	2	42.6 (24.8 ± 73.3)	4.55 (2.47–8.37)	<0.001
NT-proBNP	2	1,874 (545 ± 6,447)	1.66 (1.05–2.67)	0.022
ST2	3	42.6 (24.8 ± 73.3)	3.98 (2.15–7.94)	<0.001
NT-proBNP	3	1,874 (545 ± 6,447)	1.85 (1.02–3.45)	0.046

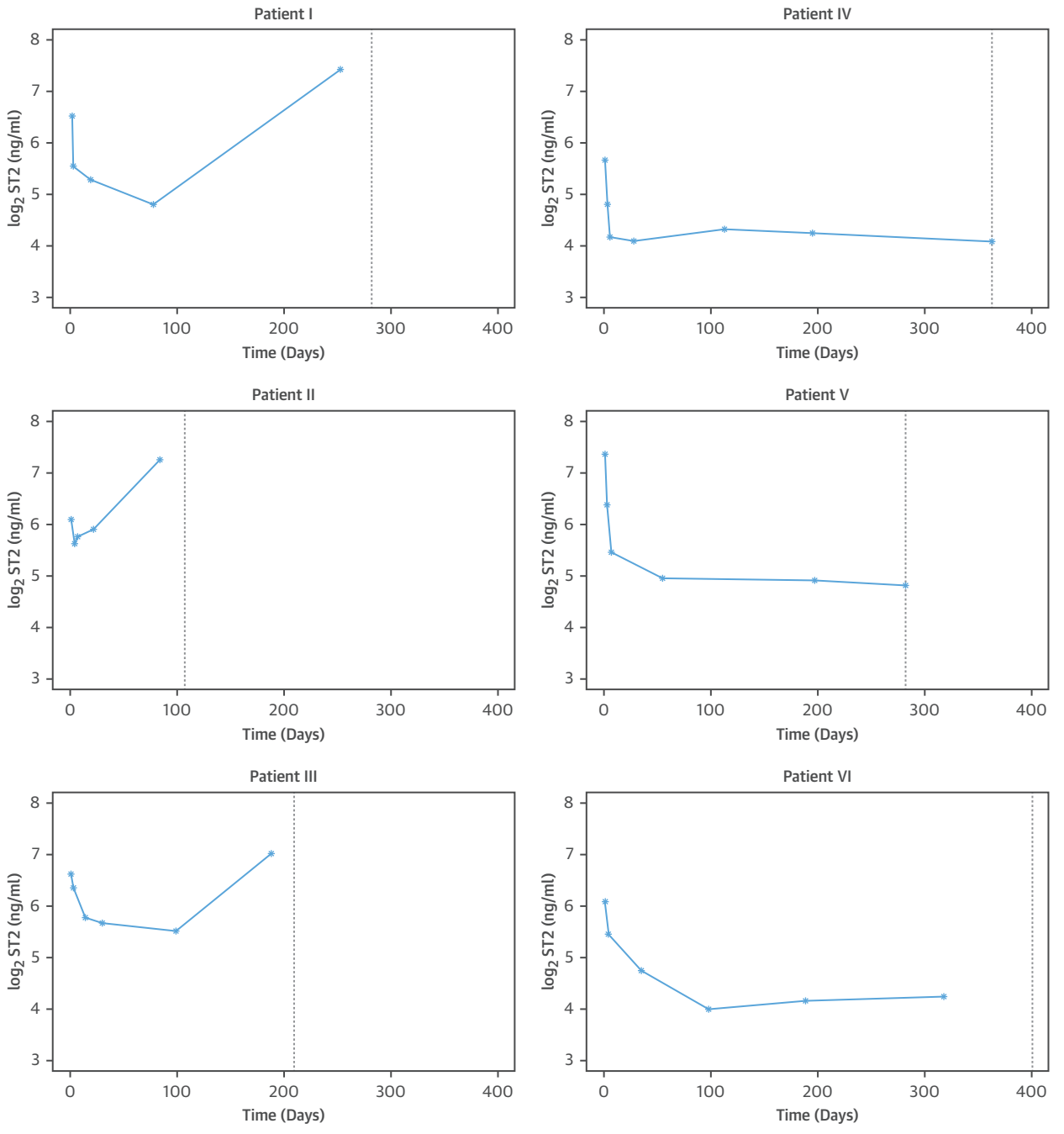
*Model 1 unadjusted; model 2 adjusted for repeated measurements of NT-proBNP or ST2, age, and sex; model 3 adjusted for repeated measurements of NT-proBNP or ST2, age, sex, systolic blood pressure, diabetes mellitus, left ventricular ejection fraction, previous hospitalization for HF during the last 6 months, ischemic HF, body mass index, eGFR, and use of medication at hospital discharge (angiotensin-converting enzyme inhibitor and/or angiotensin II receptor antagonist, beta-blocker, diuretics). †Mean ± 1 SD of the patient-specific geometric mean biomarker level during follow-up (presented on the linear scale). ‡Hazard ratios are related to a 1 SD increase of biomarker level (on the log scale) at any point in time.

Abbreviations as in Tables 1 and 2.

Figure 1 shows the measured ST2 levels of 3 individuals who had a U-shaped ST2 pattern and of 3 individuals who had a J-shaped pattern. Of the patients who reached the primary endpoint, 56% had a U-shaped ST2 pattern preceding the occurrence of the endpoint event, as illustrated in Figure 1 patients I, II, and III. Figure 1 patients IV, V, and VI are examples of J-shaped ST2 patterns in patients who did not reach the primary endpoint. When a J-shaped ST2 pattern was present during follow-up, 82% of the patients remained event free.

Figure 2 and the Central Illustration show the average estimated biomarker level and the individual biomarker measurements in patients with and without the primary endpoint adjusted according to model 3. During initial hospitalization, when all patients were treated for decompensated HF, the average estimated ST2 level decreased (Figure 2A).

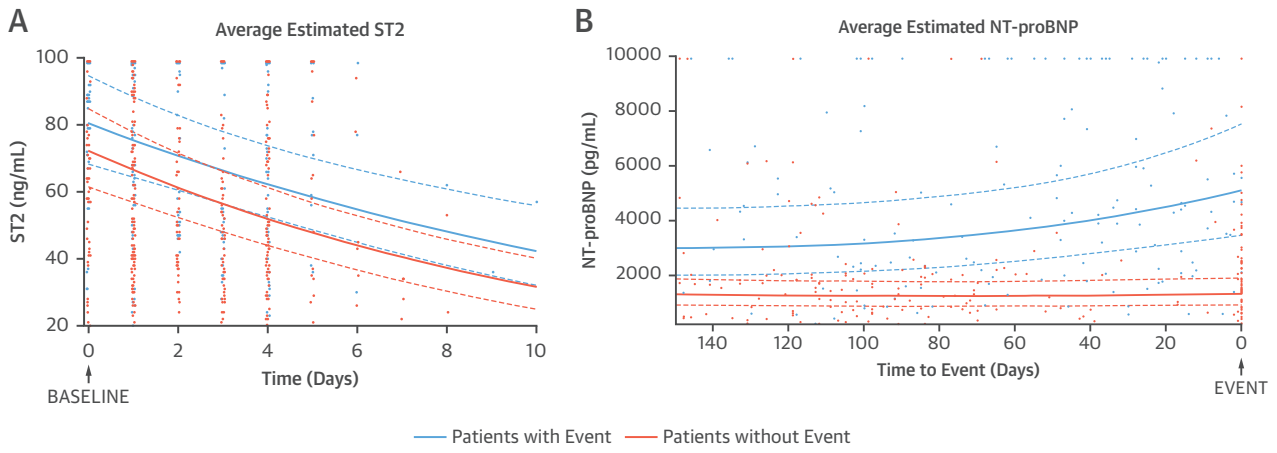
FIGURE 1 Examples of the ST2 Pattern During Follow-Up in Different Patients



The ST2 level of 6 patients during follow-up. The **vertical dotted line** represents the time of occurrence of the primary endpoint or the scheduled end of follow-up. Patients I, II, and III demonstrate a U-shaped ST2 pattern and reach the primary endpoint. Patients IV, V, and VI demonstrate a J-shaped ST2 pattern and remained event free during follow-up.

Following initial hospitalization, the average estimated ST2 levels in patients who reached the primary endpoint were higher than in their counterparts who remained free of the primary endpoint. Furthermore, the average estimated ST2 levels increased several weeks before the time of the primary endpoint (**Central Illustration**). The shape of the average estimated NT-proBNP pattern following initial

FIGURE 2 Average Estimated Biomarker Pattern, Combined With Individual Biomarker Measurements, During Follow-Up in Patients With and Without the Primary Endpoint



(A) Average estimated ST2 pattern during initial hospitalization for decompensated heart failure for patients with and without the primary endpoint. **(B)** Average estimated N-terminal pro–B-type natriuretic peptide (NT-proBNP) pattern before the primary endpoint or at the end of follow-up for patients with and without the primary endpoint. The average estimated ST2 and NT-proBNP levels are adjusted for age, sex, systolic blood pressure, diabetes mellitus, left ventricular ejection fraction, previous hospitalization for heart failure during the last 6 months, ischemic heart failure, body mass index, estimated glomerular filtration rate, and use of medication at hospital discharge (angiotensin-converting enzyme inhibitor and/or angiotensin II receptor antagonist, beta-blocker, diuretics) (model 3).

hospitalization was comparable to that of the average estimated ST2 pattern (Figure 2B).

DISCUSSION

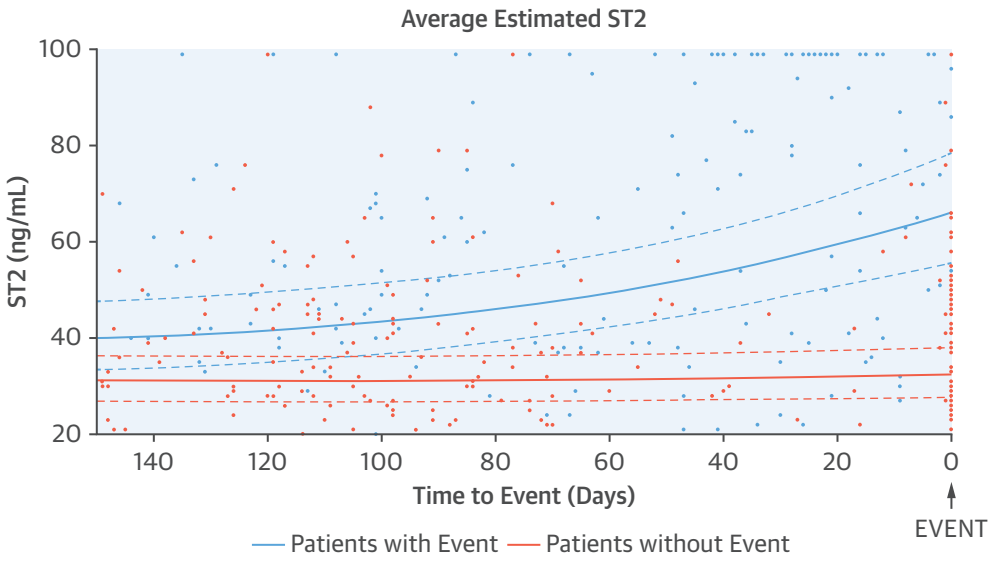
This study clearly demonstrates that baseline ST2 levels, and especially repeated ST2 measurements, are a strong and independent predictor of the composite endpoint of all-cause mortality or readmission for HF during 1-year follow-up in patients admitted with acute HF. Our results support the concept that serial measurements of ST2 offer substantial incremental prognostic value to (repeatedly measured) NT-proBNP, which is still considered the gold standard biomarker in HF.

The TRIUMPH study was designed to identify and validate novel biomarkers to improve prognostication in HF. TRIUMPH was designed as a translational study program combining biological discovery of novel biomarkers, technological advances, and clinical validation in patients presenting with acute HF. In the clinical validation study, the biomarkers were evaluated for their prognostic properties by using a unique design of repeated measurements during 1-year follow-up. Within TRIUMPH, ST2 was labeled as a biomarker with high potential for improving prognostication.

It has been established that ST2 levels in patients with acutely decompensated HF are useful for prognostication (3,23,24). Our observation that baseline ST2 level was significantly associated with all of the predefined study endpoints confirms this. In line with previous studies, the association between baseline ST2 level and readmission for HF is weaker than the association between baseline ST2 and the mortality endpoints when adjusted for all potential confounders and baseline NT-proBNP.

Repeated ST2 measurements were strongly related to the primary endpoint, as well as its separate components. The association between repeated ST2 level and the primary endpoint was highly significant and considerably stronger than the association between baseline ST2 level and the primary endpoint. Repeated measurements take into account the dynamic and continuous change in ST2 level over time that may better reflect the true changes that occur in the underlying pathophysiological processes in the individual patient with HF. In this study, repeated ST2 measurements were used to estimate the instantaneous ST2 levels (i.e., the estimated ST2 level at any point in time during the follow-up period). These estimated instantaneous ST2 levels were strongly associated with the occurrence of the predefined endpoints, most likely because the level of the estimated ST2 level is close to the true ST2 level and therefore reflects the true cardiac condition of the patient at that point in time during follow-up. This is important because HF is a dynamic and often

CENTRAL ILLUSTRATION **Average Estimated ST2 Patterns, Combined With Individual ST2 Measurements**



van Vark, L.C. et al. J Am Coll Cardiol. 2017;70(19):2378–88.

Average estimated ST2 patterns, combined with individual ST2 measurements, before the primary endpoint (composite of all-cause mortality and heart failure rehospitalization) or end of follow-up. Separate graphs are shown for patients who experienced the primary endpoint **(blue)** and those who did not **(orange).** The average estimated ST2 levels are adjusted for age, sex, systolic blood pressure, diabetes mellitus, left ventricular ejection fraction, previous hospitalization for heart failure during the last 6 months, ischemic heart failure, body mass index, estimated glomerular filtration rate, use of medication at hospital discharge (angiotensin-converting enzyme inhibitor and/or angiotensin II receptor antagonist, beta-blocker, diuretics), and N-terminal pro-B-type natriuretic peptide.

progressive disease in which inflammation, cardiac fibrosis, and remodeling are ongoing processes that cannot be captured in a single biomarker assessment at 1 point in time (5).

Another finding of the present study is that the estimated average ST2 levels increase in patients before the primary endpoint is reached, whereas the average estimated ST2 level in patients without the primary endpoint during follow-up stabilizes. The slope of the ST2 trajectory itself did not add significant prognostic information to the estimated instantaneous ST2 level. An explanation for this finding could be that the distribution of the biomarker measurements is not ideal for assessment of the instantaneous slope. To clarify these findings, a post hoc analysis was performed to define the ST2 pattern in individual patients. This analysis demonstrated that almost twice as many patients who reached the primary endpoint during follow-up had a so-called U-shaped ST2 pattern, compared with patients without an event. Furthermore, when a J-shaped ST2 pattern was identified, 82% of these patients remained event free during 1 year of follow-up. Although we acknowledge that the classification of the ST2 pattern may be affected by subjectivity and that one should be careful about drawing conclusions from this post hoc analyses, these findings suggest that the progression of ST2 levels may be important for the evaluation of an HF patient. The increase or stabilization of the ST2 level may be a useful variable in daily practice not only for stratifying patients in high-risk and low-risk categories but even more so for acting on an anticipated cardiac deterioration of a patient when ST2 levels rise during outpatient clinic follow-up visits.

Another important finding of the present study is that repeated ST2 measurements conferred independent prognostic information in addition to that offered by repeated NT-proBNP measurements. The finding that NT-proBNP and ST2 levels reflect different underlying pathophysiological processes in HF may be the most important reason for this observation. NT-proBNP is a marker of volume overload (25). ST2 responds to mechanical overload as well, but it is also a

marker of cardiac fibrosis, inflammation, and remodeling (8). In this way, ST2 and NT-proBNP levels provide complementary information on the pathophysiological state, as well as information relevant to the assessment of prognosis. With respect to prognostication in HF, the results of the present study therefore provide evidence not only for the use of repeated ST2 measurements, but also for the combined use with (repeatedly measured) NT-proBNP levels.

This study combined repeated ST2 measurements with repeated NT-proBNP measurements in patients with acute HF and therefore adds important evidence to the statement in the AHA/ACC guidelines for management of HF that ST2 is considered useful for prognostication and therapy monitoring, in addition to the use of NT-proBNP (19).

Future studies should assess the value of repeated ST2 measurements when used to guide treatment decisions. It may be hypothesized that treatment should be intensified in patients with high ST2 levels or unfavorable (increasing) ST2 patterns. Moreover, repeated ST2 measurements may be helpful to identify patients who are more likely to respond to certain treatments. Additional studies should also determine the number of ST2 measurements needed for optimal prognostication and therapy monitoring. The frequency by which ST2 levels should be measured may not be identical for each patient, but they may depend on the clinical condition of the patient, the treatment given, the ST2 level, and the progression of ST2 levels during follow-up. On the basis of these factors, an individual survival curve could be plotted, which should be used for planning of the next ST2 measurement. Because of the significantly lower biological variability of ST2 compared with NT-proBNP in patients with stable HF, it has been suggested that ST2 may be a better biomarker for monitoring patients with HF (26).

STUDY LIMITATIONS. Although this study is a large multicenter prospective observational study, it seems that the studied population is not completely representable for the average HF population. The mean age in our study population is 74 years, and women are underrepresented. Moreover, only 17% of the included patients with HF have a preserved ejection fraction. Future studies need to investigate whether similar results are found in a population that represents more women, different age groups, and HF patients with a preserved ejection fraction.

CONCLUSIONS

The TRIUMPH study clearly demonstrates that repeated measurements of ST2 are a strong and independent predictor of adverse outcome in patients following admission for acute HF. The repeated ST2 measurements identified patients at a substantially higher risk of adverse events than did baseline ST2 levels alone. In addition, repeated ST2 measurements offer incremental prognostic value to that conferred by other known risk factors and, importantly, repeated measurements of NT-proBNP. These results suggest that repeated ST2 measurements in addition to NT-proBNP measurements may be helpful in clinical practice to identify patients with HF who are at increased risk of adverse outcomes.

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PERSPECTIVES

COMPETENCY IN MEDICAL KNOWLEDGE: During the first year after hospitalization with acute HF, rising or persistently elevated blood levels of the interleukin receptor ST2 correlate with adverse clinical outcomes, including readmission for decompensated HF and all-cause mortality. Repeated measurements of ST2 have prognostic implications beyond those conferred by levels of NT-proBNP.

TRANSLATIONAL OUTLOOK: Future studies should assess the utility of serial measurements of ST2 levels to guide specific therapeutic interventions during long-term management of patients with chronic HF.

REFERENCES

1. Go AS, Mozaffarian D, Roger VL, et al. Heart disease and stroke statistics—2014 update: a report from the American Heart Association. Circulation 2014;129:e28–292.

2. Manzano-Fernandez S, Mueller T, Pascual-Figal D, Truong QA, Januzzi JL. Usefulness of soluble concentrations of interleukin family member ST2 as predictor of mortality in patients with acutely decompensated heart failure relative to left ventricular ejection fraction. Am J Cardiol 2011;107:259–67.

3. Lassus J, Gayat E, Mueller C, et al. Incremental value of biomarkers to clinical variables for mortality prediction in acutely decompensated heart failure: the Multinational Observational Cohort on Acute Heart Failure (MOCA) study. Int J Cardiol 2013;168:2186–94.

4. Januzzi JL Jr., Sakhuja R, O'Donoghue M, et al. Utility of amino-terminal pro-brain natriuretic peptide testing for prediction of 1-year mortality in patients with dyspnea treated in the emergency department. Arch Intern Med 2006;166:315–20.

5. Braunwald E. Heart failure. J Am Coll Cardiol HF 2013;1:1–20.

6. Schmitz J, Owyang A, Oldham E, et al. IL-33, an interleukin-1-like cytokine that signals via the IL-1 receptor-related protein ST2 and induces T helper type 2-associated cytokines. Immunity 2005;23:479–90.

7. Sanada S, Hakuno D, Higgins LJ, Schreiter ER, McKenzie AN, Lee RT. IL-33 and ST2 comprise a critical biomechanically induced and cardioprotective signaling system. J Clin Invest 2007;117:1538–49.

8. Weinberg EO, Shimpo M, De Keulenaer GW, et al. Expression and regulation of ST2, an interleukin-1 receptor family member, in cardiomyocytes and myocardial infarction. Circulation 2002;106:2961–6.

9. Weinberg EO, Shimpo M, Hurwitz S, Tominaga S, Rouleau JL, Lee RT. Identification of serum soluble ST2 receptor as a novel heart failure biomarker. Circulation 2003;107:721–6.

10. Bartunek J, Delrue L, Van Durme F, et al. Nonmyocardial production of ST2 protein in human hypertrophy and failure is related to diastolic load. J Am Coll Cardiol 2008;52:2166–74.

11. Ky B, French B, McCloskey K, et al. High-sensitivity ST2 for prediction of adverse outcomes in chronic heart failure. Circ Heart Fail 2011;4:180–7.

12. Aimo A, Vergaro G, Passino C, et al. Prognostic value of soluble suppression of tumorigenicity-2 in chronic heart failure: a meta-analysis. J Am Coll Cardiol HF 2017;5:280–6.

13. Boisot S, Beede J, Isakson S, et al. Serial sampling of ST2 predicts 90-day mortality following destabilized heart failure. J Card Fail 2008;14:732–8.

14. Gaggin HK, Szymonifka J, Bhardwaj A, et al. Head-to-head comparison of serial soluble ST2, growth differentiation factor-15, and highly-sensitive troponin T measurements in patients with chronic heart failure. J Am Coll Cardiol HF 2014;2:65–72.

15. Anand IS, Rector TS, Kuskowski M, Snider J, Cohn JN. Prognostic value of soluble ST2 in the Valsartan Heart Failure Trial. Circ Heart Fail 2014;7:418–26.

16. Tang WH, Wu Y, Grodin JL, et al. Prognostic value of baseline and changes in circulating soluble ST2 Levels and the effects of nesiritide in acute decompensated heart failure. J Am Coll Cardiol HF 2016;4:68–77.

17. Broch K, Ueland T, Nymo SH, et al. Soluble ST2 is associated with adverse outcome in patients with heart failure of ischaemic aetiology. Eur J Heart Fail 2012;14:268–77.

18. Demissei BG, Cotter G, Prescott MF, et al. A multimarker multi-time point-based risk stratification strategy in acute heart failure: results from the RELAX-AHF trial. Eur J Heart Fail 2017;19:1001–10.

19. Yancy CW, Jessup M, Bozkurt B, et al. 2013 ACCF/AHA guideline for the management of heart failure: a report of the American College of Cardiology Foundation/American Heart Association Task Force on Practice Guidelines. J Am Coll Cardiol 2013;62:e147–239.

20. McMurray JJ, Adamopoulos S, Anker SD, et al. ESC guidelines for the diagnosis and treatment of acute and chronic heart failure 2012: the Task Force for the Diagnosis and Treatment of Acute and Chronic Heart Failure 2012 of the European Society of Cardiology. Developed in collaboration with the Heart Failure Association (HFA) of the ESC. Eur Heart J 2012;33:1787–847.

21. Stevens LA, Coresh J, Greene T, Levey AS. Assessing kidney function—measured and estimated glomerular filtration rate. N Engl J Med 2006;354:2473–83.

22. Rizopoulos D. The R Package JMbayes for fitting joint models for longitudinal and time-to-event data using MCMC. arXiv:1404.7625 [stat.CO]. Ithaca, NY: Cornell University Library, April 30, 2014.

23. Rehman SU, Mueller T, Januzzi JL. Characteristics of the novel interleukin family biomarker ST2 in patients with acute heart failure. J Am Coll Cardiol 2008;52:1458–65.

24. Shah RV, Chen-Tournoux AA, Picard MH, van Kimmenade RR, Januzzi JL. Serum levels of the interleukin-1 receptor family member ST2, cardiac structure and function, and long-term mortality in patients with acute dyspnea. Circ Heart Fail 2009;2:311–9.

25. Hunt PJ, Richards AM, Nicholls MG, Yandle TG, Doughty RN, Espiner EA. Immunoreactive amino-terminal pro-brain natriuretic peptide (NT-PROBNP): a new marker of cardiac impairment. Clin Endocrinol (Oxf) 1997;47:287–96.

26. Piper S, deCourcey J, Sherwood R, Amin-Youssef G, McDonagh T. Biologic variability of soluble ST2 in patients with stable chronic heart failure and implications for monitoring. Am J Cardiol 2016;118:95–8.

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