ORIGINAL RESEARCH

Repeat Measurements of High Sensitivity Troponins for the Prediction of Recurrent Cardiovascular Events in Patients With Established Coronary Heart Disease: An Analysis From the KAROLA Study

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***Background*—**High-sensitivity cardiac troponins T and I (hs-cTnT and hs-cTnI) are established biomarkers for myocardial injury and used for diagnostic and prognostic purposes. However, whether repeat measurements improve prediction of recurrent cardiovascular disease (CVD) events in patients with stable coronary heart disease (CHD) after adjustment for several other novel biomarkers remains unclear.

***Methods and Results*—**We measured both troponins in 873 coronary heart disease patients from the KAROLA (Langzeiterfolge der Kardiologischen Anschlussheilbehandlung) study about 9 weeks after their initial acute event (baseline) and after 12 months, followed them for 12 years, assessed a combined CVD end point, and adjusted for several risk factors. As we found evidence for effect modification, results were stratified according to presence of myocardial infarction at baseline. During follow-up, 186 fatal and non-fatal CVD events occurred. Both baseline and 12-months troponin concentrations were significantly associated with CVD events in patients without myocardial infarction at baseline; in tendency 12 months of troponin showed stronger hazard ratios (hs-cTnT: hazard ratios 1.91 (95% CI 1.17–3.11) versus baseline values 1.71 (95% CI 1.08–2.70) and for hs-cTnI: hazard ratio 1.55 (95% CI 1.05–2.30) versus baseline value 1.22 (95% CI 0.88–1.68) in the fully and simultaneously adjusted model.

***Conclusions*—**Both troponins are consistently associated with recurrent cardiovascular events after adjustment for emerging risk factors during follow-up in our study especially evident in patients without myocardial infarction at baseline. Troponin values at 12 months of follow-up showed independent associations with future CVD events in addition to baseline assessments of troponins. **(*J Am Heart Assoc*. 2019;8:e011882. DOI: 10.1161/JAHA.118.011882.)**

Key Words: cohort study • coronary heart disease • hsTroponin I • hsTroponin T • risk prediction

Despite comprehensive efforts in terms of optimized patient care and management of individual risk profiles, recurrent cardiovascular events remain of major concern in patients with established coronary heart disease (CHD).[1] Hence, biomarkers with prognostic impact are of paramount interest to identify those patients being at high risk for future events and to identify those who might benefit from early intervention.

Cardiac troponins not only represent a cornerstone to diagnose an acute coronary syndrome for >15 years,[2] the introduction of high-sensitivity (hs) assays led to an even more accurate detection of lower levels, which have been shown to be associated with incident CHD in the general population[3] as well as with recurrent events in patients with stable disease.[4,5] Since CHD remains a chronic disease which is modified by multiple exogenous factors (eg, nutrition,

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Clinical Perspective

What Is New?

- We have shown that both high-sensitivity cardiac troponins T and I were associated with recurrent cardiovascular events after adjustment for emerging risk factors during 12-year follow-up in our cohort of 873 coronary heart disease patients.

What Are the Clinical Implications?

- Since the repeat measurements of high-sensitivity cardiac troponins seem to improve the risk prediction beyond single measurements taken shortly after an initial cardiovascular event, repeat troponin measurements during follow-up in this patient group may help identify patients at high-risk for recurrent events.

physical activity, medication) over a long period of time, the question arises whether repeat measurements of hs-troponins can further improve the accuracy to predict future cardiovascular events in patients already known to suffer from CHD.

Former studies suggested that repeat measurements of troponin-I, hs-cTnT, and hs-cTnI might represent valuable markers for incident CHD,[6,7] death, and heart failure in the general population.[8] In addition, a study evaluated changes of hs-cTnI over time in patients with stable CHD within a rather short interval (4 months).[9] In this regard, we sought to determine the potential use of repeat measurements at baseline and 12 months of hs-cTnT and hs-cTnI for the prediction of recurrent cardiovascular events in patients with established CHD and long-term follow-up after adjustment for numerous established and emerging biomarkers.

Methods

Because of ethical restrictions on data protection issues and the study specific consent text and procedure, the data cannot be made publicly available, but data are available to all interested researchers upon request.

Study Population

The KAROLA (Langzeiterfolge der Kardiologischen Anschlussheilbehandlung) study was initiated to elucidate the long-term course of patients who either experienced an acute cardiovascular event (eg, myocardial infarction [MI]) or underwent elective coronary artery bypass grafting (CABG) or percutaneous coronary intervention (PCI) between January 1999 and May 2000. Patients (aged 30 to 70 years, meeting the *International Classification of Diseases, Ninth Revision (ICD-9)* criteria [codes 410–414] for CHD) suffering from one of the above mentioned conditions for the first time took part in a 3-week rehabilitation program in 2 collaborating rehabilitation centers in Germany (*Schwabenland-Klinik*, Isny, and *Klinik im Südpark*, Bad Nauheim).[5] This in-hospital rehabilitation program was initiated up to 3 weeks after the acute event. All patients who were eligible for enrollment gave written informed consent to participate in this study. The ethics committees of the Universities of Ulm, Ulm and Heidelberg, Heidelberg, and the ethics boards of the chambers of physicians of the federal states of Hessen, Frankfurt, and Baden-Wuerttemberg, Stuttgart, approved the study.[10,11]

Data Collection

All patients were asked to complete a comprehensive questionnaire on sociodemographic factors and lifestyle changes and were invited to participate in an active follow-up at 1, 3, 4.5, 6, 8, 10, and 13 years after the initial evaluation during their 3-week rehabilitation program. In addition, information was taken from the patient`s hospital charts. Data on left ventricular function were collected from medical charts and were documented on a 4-point semiquantitative scale as normal (ejection fraction [EF] >65%), mild depression (EF 50% to ≤65%), moderate depression (EF 35% to ≤50%), or severe depression (EF <35%). Frequency of leisure time physical activity was assessed with the question "On average, to what extent (hours per week) have you engaged in physically strenuous and sweat-inducing activity in your leisure time in the past 12 months (eg, cycling, speedy hiking, gardening, sport) (please do not consider periods of hospital stays)?" Answers were categorized into "none to 2 h/week", "3 to 4 h/week", "5 to 8 h/week", "≥9 h/week". Further information during follow-up on medication, treatment, diagnoses, and potential secondary cardiovascular events was provided by the primary care physician.

In case a participant had died during follow-up, the underlying cause was obtained from the official death certificate, which can be accessed at the local health authorities, and was documented according to *ICD-9* codes 390–459 and *ICD-10* codes I0–I99 and R.57.0. As pointed out earlier,[5,10,11] secondary cardiovascular events were defined as the following: CVD as the main cause of death and non-fatal stroke or myocardial infarction as reported by the primary care physician. Since 28 out of the initial 901 individuals experienced CVD events within the first 12 months, we excluded them from further analyses.

Laboratory Measurements

At the time of discharge from the rehabilitation clinic (about 9 weeks after their initial acute event), fasting baseline blood samples were taken and were stored at −80°C for future analyses. The 12-months serum samples were obtained from

the primary care physician and sent to the study center and consecutively stored at −80°C. A high-sensitivity (hs) assay by Roche Diagnostics on an Elecsys 2010 platform (Roche) was run for all hs-cTnT (high-sensitivity cardiac troponin T) measurements with a lower limit of detection (LoD) of 5 ng/L and inter-assay coefficient of variation of 3.6% and 2.9% at concentrations of 42 and 2.82 ng/L, respectively. In parallel, hs-cTnI was determined on an Abbott ARCHITECT i1000 platform with an LoD of 2.0 ng/L and inter-assays of coefficient of variation being between 5.9 and 6.7% at 3 different concentrations.[5]

Statistical Methods

Continuous variables for the sociodemographic and medical characteristics were presented as means and standard deviations, while discrete variables were presented as numbers and percentages.[5] We described the association between both troponin subtypes (difference between log-transformed values between 1 year follow-up and baseline) and various covariates by a Kruskal–Wallis test and in addition, by using age- and sex-adjusted partial Spearman rank correlation coefficients. A Cox proportional hazards regression analysis was used to evaluate the prognostic value of both troponin subtypes at baseline and at 12-month follow-up for secondary cardiovascular events. For such analyses the troponin levels were natural log transformed (ln)[12]; values <LoD were set at LoD divided by 2. Start of observation time was after conduct of 1 year follow-up and therefore events which occurred within the first year were excluded. After adjustment for age and sex we included multiple risk factors such as body mass index (BMI), smoking status (never, current, former smoker), duration of school education (<10 years, ≥10 years), hospital site (Isny, Bad Nauheim), family status (married, other), history of MI (yes, no), history of hypertension (yes, no), history of diabetes mellitus (yes, no), severity of CHD (number of affected epicardial coronary vessels at baseline), initial management of CHD (percutaneous coronary intervention [yes, no]), intake of ß-blockers (yes, no), intake of angiotensin-converting enzyme (ACE) inhibitors (yes, no), intake of diuretics (yes, no), intake of lipid-lowering drugs (yes, no), high-density lipoprotein and low-density lipoprotein cholesterol, and for estimated glomerular filtration rate (CKD-EPI cystatin C equation). To avoid over-adjustment, we excluded those factors, which had neither statistically significant associations with the occurrence of secondary events (at an α-level of 0.1) nor changed hs-cTnT or hs-cTnI point estimates, respectively, in a relevant way (ie, by >10%) in a manual backward selection. At the end multivariable adjustment included the following variables: age, sex, BMI, systolic blood pressure, left ventricular function, smoking status, diabetes mellitus, total cholesterol, HDL cholesterol, CKD Epi (cystatin C-based), and use of statins. For the simultaneous adjusted models we assessed multicollinearity by examining tolerance and the variance inflation factor. In addition, measures of discrimination and reclassification were assessed with Cox-proportional hazards models.[13,14] The net reclassification improvement by adding hs-cTnT and hs-cTnI as well as both markers simultaneously to a basic model was calculated according to the risk strata of <10%, 10% to 20%, and >20% of predicted probability for a cardiovascular event. The individual relative 12-month change of each hs-cTN has been defined as "12-months ln hs-cTn minus baseline ln hs-cTn divided by baseline ln cTn multiplied by 100".[12] The associations of these relative changes and the incidence of secondary cardiovascular events were evaluated by using Cox proportional hazards models in which these relative changes were entered either as continuous variable or in categories. Associations were adjusted for baseline levels of ln hs-cTnT and ln hs-cTnI, age and sex, BMI, blood pressure, left ventricular function, smoking, history of diabetes mellitus, total cholesterol, HDL cholesterol, standardized estimated glomerular filtration rate, and use of statins. The categorical analysis was done by including those with a 12-months increase of troponin levels into the upper category; those with a 12-months decrease of troponin levels were divided into tertiles. As we found evidence of effect modification (eg, P=0.007 for product term of ln-transformed hs-cTnT and myocardial infarction [MI] at baseline) we stratified results also accordingly. All statistical procedures were performed with the SAS statistical software package (release 9.4 SAS Institute Inc.).

Results

Demographic, Clinical, and Laboratory Characteristics

Initially, baseline, and 12-month troponin values were obtained in 901 patients. The sociodemographic, clinical, and laboratory characteristics of the remaining 873 patients with clinically manifest CHD are presented in Table 1 in total and stratified according to subsequent CVD-event. In total the mean age of the patients was 58.9 years while most were men (85.2%). Traditional risk factors observed were smoking in 67.4% (former and current smokers), history of diabetes mellitus in 16%, and a history of hypertension in 56.4%. With respect to the main analyses, the median of hs-cTnT and hs-cTnI levels at baseline were 13.8 and 14.0 ng/L, respectively. At 12 months of follow-up the mean concentrations decreased to 7.9 and 8.1 ng/L. The correlation coefficient between hs-cTnT and hs-cTnI at baseline was 0.62 and at 1-year follow-up 0.57, respectively.

As we found effect modification between troponins and MI at baseline we stratified results also accordingly. Patients with

Table 1. Sociodemographic, Clinical, and Laboratory Characteristics in 873 Patients With Clinically Manifest Coronary Heart Disease

Characteristics at Baseline	Total	With Subsequent CVD Event	Without Subsequent CVD Event
Age, y (μ, SD)	58.9±8.1	61.2±7.0	58.3±8.3
Men, n (%)	744 (85.2)	162 (87.1)	582 (84.7)
Myocardial infarction, n (%)	444 (50.9)	95 (51.1)	349 (50.8)
History of heart failure, n (%)	103 (12.2)	28 (15.5)	75 (11.3)
Clinical score (angiographic evaluation), n (%)			
1 vessel disease	215 (24.6)	35 (18.8)	180 (26.2)
2 vessel disease	240 (27.5)	49 (26.3)	191 (27.8)
3 vessel disease	370 (42.4)	91 (48.9)	279 (40.6)
Unknown	36 (4.1)	10 (5.4)	26 (3.8)
School education <10 years, n (%)	513 (58.8)	119 (64.0)	394 (57.4)
Body mass index, kg/m^2 (μ, SD)	26.8±3.2	27.3±3.5	26.7±3.1
Smoking status, n (%)			
Never	285 (32.7)	59 (31.7)	226 (32.9)
Ex	549 (62.9)	117 (62.9)	432 (62.9)
Current	39 (4.5)	10 (5.4)	29 (4.2)
History of diabetes mellitus, n (%)	140 (16.0)	43 (23.1)	97 (14.1)
History of hypertension, n (%)	492 (56.4)	117 (62.9)	375 (54.6)
PCI, n (%)	336 (38.7)	70 (38.0)	266 (38.8)
CABG, n (%)	426 (49.0)	95 (51.6)	331 (48.3)
Lipid-lowering agents, n (%)	671 (77.0)	134 (72.0)	537 (78.4)
ASA, n (%)	769 (88.3)	159 (85.5)	610 (89.1)
Clopidogrel or Ticlopidine, n (%)	75 (8.6)	15 (8.1)	60 (8.7)
Beta-blockers, n (%)	765 (87.8)	165 (88.7)	600 (87.6)
ACE-inhibitors, n (%)	443 (50.9)	112 (60.2)	331 (48.3)
Total cholesterol, mg/dL (μ, SD)	167.9 (32.1)	169.3 (33.3)	167.6 (31.8)
LDL-cholesterol, mg/dL (μ, SD)	100.1 (29.1)	101.1 (29.6)	99.9 (29.0)
HDL-cholesterol, mg/dL (μ, SD)	39.5 (10.5)	38.6 (10.1)	39.8 (10.6)
C-reactive protein, mg/L*	3.28 (1.22; 8.28)	3.76 (1.43; 9.76)	3.16 (1.17; 7.77)
Cystatin C, mg/L*	1.03 (0.93; 1.17)	1.11 (0.97; 1.31)	1.02 (0.93; 1.13)
NT-proBNP, pg/mL*	547.60 (276.50; 1049.50)	935.50 (420.80; 1663.00)	486.40 (248.60; 885.40)
MR-proANP, pmol/L*	135.65 (95.34; 180.65)	168.10 (118.90; 234.70)	128.80 (92.01; 171.20)
sST2, ng/mL*	28.76 (23.88; 34.68)	29.70 (24.52; 35.70)	28.45 (23.82; 34.26)
GDF15, ng/L	1215.0 (924.0; 1619.5)	1423.5 (1101.0; 2114.0)	1149 (891; 1526)
Galectin 3, ng/mL	12.8 (10.6; 15.7)	13.7 (11.5; 16.6)	12.5 (10.5; 15.3)
hs-cTnT* (ng/L) baseline	13.80 (8.87; 21.30)	17.20 (11.30; 27.40)	12.80 (8.25; 19.30)
hs-cTnT* (ng/L) 1 YFU	7.89 (5.44; 11.60)	10.50 (7.21; 15.20)	7.45 (5.16; 10.50)
hs-cTnI* (ng/L) baseline	4.0 (9.3; 23.0)	18.9 (12.4; 29.0)	13.2 (8.9; 21.2)
hs-cTnI* (ng/L) 1 YFU	8.1 (5.7; 12.0)	11.95 (7.10; 18.90)	7.5 (5.5; 10.6)
hs-cTnT >5 ng/L baseline, n (%)	817 (93.6)	182 (97.9)	635 (92.4)
hs-cTnT >5 ng/L 1 YFU, n (%)	691 (79.2)	164 (88.2)	527 (76.7)
hs-cTnT >14 ng/L baseline, n (%)	430 (49.3)	119 (64.0)	311 (45.3)

Continued

Table 1. Continued

Characteristics at Baseline	Total	With Subsequent CVD Event	Without Subsequent CVD Event
hs-cTnT >14 ng/L 1 YFU, n (%)	150 (17.2)	59 (31.7)	91 (13.3)
hs-cTnI >2 ng/L baseline, n (%)	873 (100)	186 (100)	687 (100)
hs-cTnI >2 ng/L 1 YFU, n (%)	872 (99.9)	186 (100)	686 (99.9)
hs-cTnI >6 ng/L baseline, n (%)	832 (95.3)	181 (97.3)	651 (94.8)
hs-cTnI >6 ng/L 1 YFU, n (%)	622 (71.3)	158 (85.0)	464 (67.5)

ASA indicates acetylsalicylic acid; CABG, coronary artery bypass graft; GDF15, growth differentiation factor 15; hs-cTnT, high-sensitivity cardiac troponin T; hs-cTnI, high-sensitivity cardiac troponin I; MR-proANP, mid-regional pro-atrial natriuretic peptide; NT-proBNP, n-terminal pro-brain natriuretic peptide; PCI, percutaneous coronary intervention; sST2, soluble suppression of tumorigenicity 2.
*Median, 25th and 75th quantile cut point; 1 YFU=1-year follow-up.

baseline-MI were younger (57.3 years, SD 8.8 versus 60.6 years, SD 6.9). In addition, 20% of the patients with baseline-MI had received a coronary artery bypass grafting and 55% had received a percutaneous coronary intervention compared with 79% and 22% in the CHD group. They also had a slightly higher age-adjusted CVD mortality rate (24.1 [95% CI 19.7–29.5] versus 20.7 [95% CI 16.6–25.8]) but a somewhat lower age-adjusted general mortality rate 14.1 (95% 11.1–18.0) versus 16.1 (95% CI 12.7–20.3) (all rates per 1000 patient-years).

Figure shows these differences between 1-year follow-up values and baseline graphically. The scatter plots (Figure—Panels A and B) clearly display the significant decreases of log-transformed values of hs-cTnT and hs-cTnI ($P<0.0001$, $r=-0.53$ and $P>0.0001$, $r=-0.50$) between baseline and 1-year follow-up. In men the median of hs-cTnT and hs-cTnI levels at baseline were 14.2 and 14.0 ng/L, respectively; in women the median levels were 11.7 and 14.0 ng/L. At 12 months of follow-up the median concentrations decreased to 8.1 and 8.3 ng/L in men and to 7.2 and 6.6 ng/L in women (data not in table).

Table 2 displays the absolute changes of both troponin subtypes from baseline to 12-month follow-up (expressed as difference of log-transformed values between 1-year follow-up and baseline) according to sociodemographic characteristics and cardiovascular risk factors. Whereas the decrease of hs-cTnT did not differ between men and women, hs-cTnI decreased significantly more in female than in male patients (−0.70 versus −0.51, $P=0.011$) during follow-up. Also, a lower BMI (<25 kg/m^2) went along with more profound decrease of both troponin subtypes. While smoking habits and physical (in-)activity were not associated with changes in troponin levels, both diabetes mellitus and hypertension were associated with a significantly weaker decrease in hs-cTnT and hs-cTnI. Moreover, use of statins at baseline went along with a less strong reduction of hs-cTnT, while there was no statistically significant difference in hs-cTnI values. In terms of the initial CHD treatment, percutaneous coronary intervention treatment was associated with a weaker decrease of hs-cTnT, but vice versa with hs-cTnI. In addition, patients undergoing coronary artery bypass grafting showed a stronger decrease of hs-cTnT, but a weaker with hs-cTnI at 12 months of follow-up. Table S1 shows results stratified to baseline MI.

Table 3 provides the age- and sex-adjusted partial Spearman rank correlation coefficients (r) between established and emerging risk factors of CHD at baseline and absolute changes of troponins. Emerging risk factors were more likely to be correlated significantly with the absolute change of hs-cTnT during follow-up (eg C-reactive protein, interleukin-6, cystatin C, estimated glomerular filtration rate, GDF-15 and MR-proANP) compared with hs-cTnI. Only NT-proBNP and creatinine correlated with absolute changes of both, hs-cTnT and hs-cTnI. While looking at traditional risk factors, HDL cholesterol showed a negative correlation with changes in hs-cTnI, but not in hs-cTnT. Table S2 shows results stratified to baseline MI.

Association of hs-Troponins With Fatal and Non-Fatal CVD Events

Tables 4 and 5 quantify the association of log-transformed hs-troponins with fatal and non-fatal CVD events after adjustment for age and sex on the one hand, and with other established and relevant risk factors on the other hand stratified according to presence of baseline MI (versus coronary heart disease). During follow-up (median follow-up of 11.6 years) 186 CVD events occurred. Focusing on hs-cTnT, both measurements (either at baseline or at 12-month follow-up) were significantly associated with future fatal and non-fatal CVD events in patients with and without baseline MI. This association persisted even after adjustment for multiple covariates in patients without baseline MI. However, in patients with baseline MI this association cannot be seen after adjustment for multiple covariates. The hs-cTnT values at 12 months of follow-up had even higher prognostic point estimates than the point estimates related to the samples drawn at baseline (hazard ratio [HR] 1.34 [95% CI 1.00–1.79] versus 1.29 [95% CI

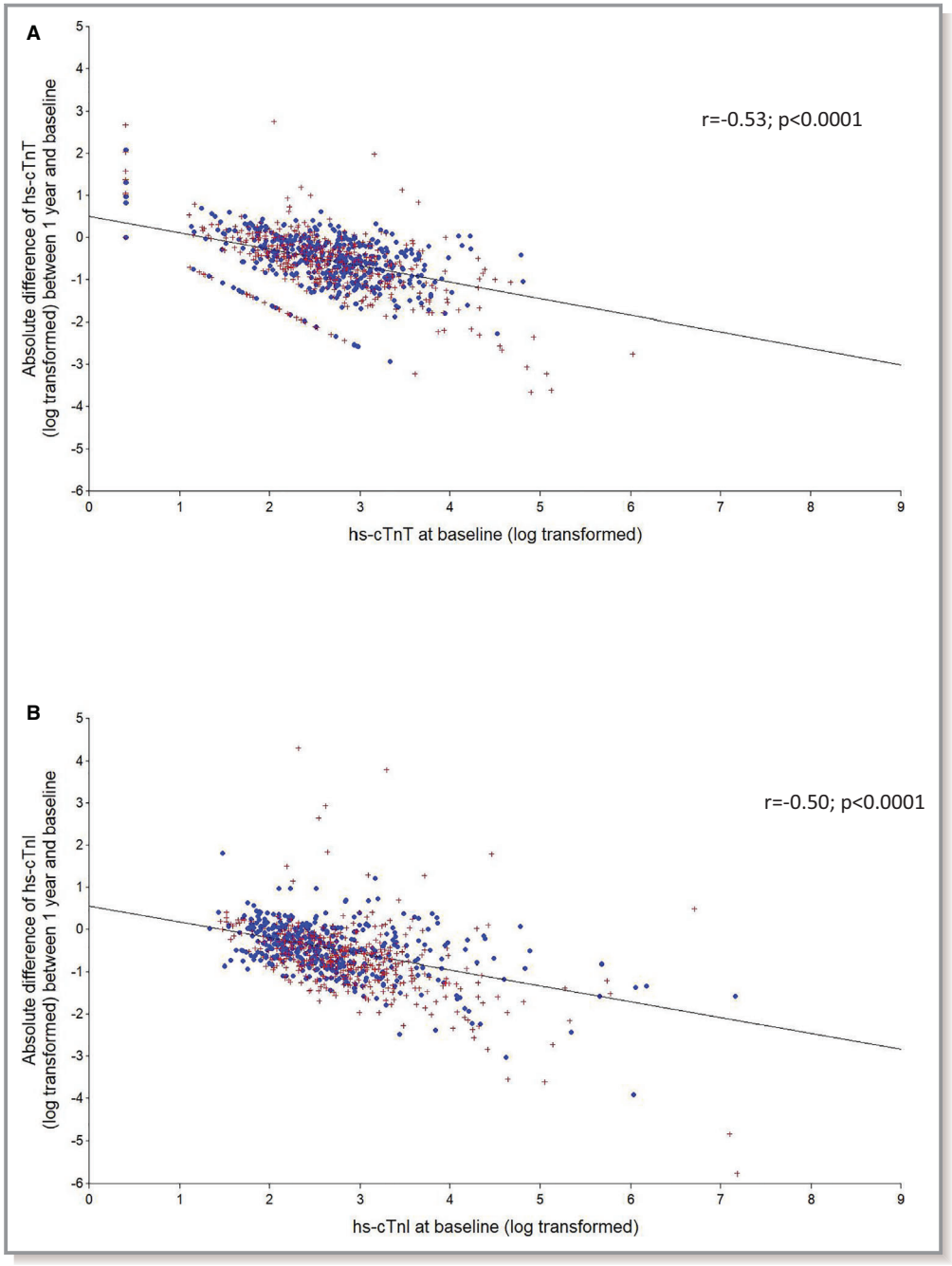

Figure. The scatter plot shows the differences between 1-year follow-up values of high-sensitivity troponin T (hs-cTnT) (**A**) and high-sensitivity cardiac troponin I (hs-cTnI) (**B**) and baseline graphically (myocardial infarction at baseline=+, coronary heart disease=•).

1.00–1.66] for patients with baseline MI and HR 3.47 [95% CI 22.43–4.97] versus 2.80 [95% CI 2.03–3.88] for patients without baseline MI, both adjusted for age and sex).

Compared with hs-cTnT, the hs-cTnI measurements in general showed smaller but still statistically significant hazard ratios for the association with subsequent CVD events in patients without baseline MI. For example, the values at 12 months of follow-up had an HR of 2.23 (95% CI 1.81–2.74) versus 1.79 (95% CI 1.52–2.12) adjusted for age and sex, $P<0.0001$. In patients with baseline MI, only the 12-month values of hs-cTnI showed a statistically significant association with subsequent CVD events. Notably if NT-proBNP was included instead of left ventricular function, it showed a strong association in both simultaneously, fully adjusted models with an HR (per unit increase after ln-transformation) of 1.48 (95% CI 1.11–1.97, $P=0.007$) for the hs-cTnT model in Table 4, and an HR of 1.63 (95% CI 1.25–2.12, $P=0.0003$) for the hs-cTnI model in Table 5.

Table 2. Absolute Change of hs-cTnT and hs-cTnI From Baseline to 1-Year Follow-Up (Median, ng/L) According to Various Sociodemographic Characteristics, and Cardiovascular Risk Factors

	n	Absolute Change of hs-cTnT Median	*P* Value	Absolute Change of hs-cTnI Median	*P* Value
Sex					
Female	129	−0.49		−0.70	
Male	744	−0.50	0.48	−0.51	0.011
Age, y					
30 to 39	21	−0.54		−0.65	
40 to 49	109	−0.69		−0.71	
50 to 59	246	−0.51		−0.51	
60 to 70	497	−0.48	0.11	−0.51	0.23
Body mass index, kg/m^2					
<25	239	−0.59		−0.60	
25 to 30	510	−0.48		−0.51	
>30	121	−0.41	0.018	−0.46	0.017
Smoking status					
Never	285	−0.50		−0.55	
Ex	549	−0.50		−0.52	
Current	39	−0.47	0.82	−0.60	0.95
Physical activity					
None to 2 h/week	282	−0.50		−0.57	
3 to 4 h/week	155	−0.47		−0.49	
5 to 8 h/week	213	−0.46		−0.51	
≥9 h/week	223	−0.51	0.75	−0.52	0.45
History of diabetes mellitus					
Yes	140	−0.36		−0.47	
No	733	−0.54	0.0028	−0.56	0.0044
History of hypertension					
Yes	492	−0.46		−0.49	
No	381	−0.55	0.0025	−0.58	0.011
Use of statins					
Yes	671	−0.46		−0.54	
No	200	−0.55	0.020	−0.52	0.88
PCI					
Yes	336	−0.43		−0.60	
No	533	−0.54	0.030	−0.49	0.020
CABG					
Yes	426	−0.57		−0.47	
No	443	−0.42	0.0001	−0.60	0.013

CABG indicates coronary artery bypass graft; hs-cTnI, high sensitivity cardiac troponin I; hs-cTnT, high sensitivity cardiac troponin T; PCI, percutaneous coronary intervention.

Relative Changes of hs-cTnT and hs-cTnI

Compared with troponin levels at baseline, a total of 730 patients showed decreasing hs-cTnT values, while only 143 patients had increasing values. In parallel, a majority of 719 patients showed lower hs-cTnI levels compared with the measurement values from baseline, while 154 patients had even higher hs-cTnI levels at 12 months. In patients without

Table 3. Age- and Sex-Adjusted Partial Spearman Rank Correlation Coefficients (*r*) Between Established and Emerging Risk Factors of Coronary Heart Disease and Absolute Change of hs-cTnT and hs-cTnI From Baseline to 1-Year Follow-Up (Median, ng/L)

	Absolute Change of hs-cTnT		Absolute Change of hs-cTnI	
	r	*P* Value	*r*	*P* Value
HDL cholesterol	−0.04	0.20	−0.13	0.0001
Total cholesterol	−0.04	0.21	−0.03	0.45
LDL cholesterol	−0.07	0.040	−0.02	0.61
Leukocytes	−0.07	0.033	0.03	0.37
C-reactive protein	−0.15	<0.0001	0.04	0.24
Interleukin-6	−0.09	0.0057	0.05	0.11
Adiponectin	−0.01	0.70	−0.07	0.031
Cystatin C	−0.10	0.0039	−0.05	0.13
eGFR (cystatin C-based)	0.10	0.0033	0.05	0.14
NT-proBNP	−0.34	<0.0001	−0.23	<0.0001
Creatinine	0.08	0.022	0.13	0.0001
HbA1c	−0.06	0.087	−0.07	0.043
MR-proANP	−0.19	<0.0001	−0.05	0.14
GDF 15	−0.15	<0.0001	−0.03	0.41
sST2	−0.04	0.28	0.05	0.12
Galectin 3	−0.04	0.20	−0.02	0.51

eGFR indicates estimated glomerular filtration rate; GDF15, growth differentiation factor 15; HbA1c, glycated hemoglobin; HDL, high-density lipoprotein; hs-cTnI, high sensitivity cardiac troponin I; hs-cTnT, high sensitivity cardiac troponin T; LDL, low-density lipoprotein; MR-proANP, mid-regional pro-atrial natriuretic peptide; sST2, soluble suppression of tumorigenicity 2.

baseline MI, a smaller decrease or even an increase went along with a higher incidence rate per 1000 person-years compared with the subjects with the highest decrease (Table 6). For example, the incidence rate was 22.4 per 1.000 patient-years for those having incremental levels of hs-cTnT compared with 15 in individuals with the greatest decrement in hs-cTnT. For hs-cTnI comparable patterns were seen (29.5 versus 19.8 per 1000 person years). Importantly, studies on the individual biological variation revealed that both short- and long-term variations of either troponins were modest.[15]

For both troponins, we observed a trend towards a monotonic increase in the hazards along the subjects without baseline MI categorized by the changes of troponin levels compared with the bottom category with the highest decrement of troponin levels within the fully adjusted model (HR for hs-cTnT: 1.73 [95% CI 0.9–3.32], 2.03 [95% CI 1.06–3.92], 3.01 [95% 1.35–6.73], *P* for trend 0.072; HR for hs-cTnI: 0.68 [95% CI 0.33–1.40], 1.25 [95% CI 0.66–2.36], 1.73 [95% CI 0.88–3.37], *P* for trend 0.079). For patients with baseline MI, no such trend was observed (see Table S3).

Discussion

The current prospective study showed that either troponins (hs-cTnT and hs-cTnI) represent a reliable predictor of recurrent cardiovascular events in our cohort consisting of 873 patients with stable CHD. Moreover, the repeat measurement of both troponin subtypes at 12 months after the inpatient rehabilitation shortly after the initial event (either acute coronary syndrome, or elective coronary artery bypass grafting) led to an even higher risk estimate for the prediction of future events, irrespective of other traditional CHD risk factors. Notably, the associations were especially evident in patients with CHD and without a baseline MI. Hence, repeat measurements of hs-troponins in these patients might help to identify those patients being at higher risk for recurrent CVD events. Such patients might benefit from an even stricter control of traditional risk factors and a closer outpatient follow-up. Our analyses showed only mild differences between the troponin subtypes on their prognostic impact and with respect to the local availability of different troponin assays, a general advice which troponin to use, cannot not be made, based on our data as HRs and clinical performance values were similiar.

Several reports showed that troponins (measured by conventional or high-sensitivity assays) are associated with future cardiovascular events both, in the general population,[6,7] as well as in patients with stable CHD.[4,5] In addition, repeat measurements of troponins in the general population elucidated the potential to mirror the predictive value of these biomarkers over time. For example, McEvoy et al reported the association of a 6-year change in hs-cTnT with incident CHD, heart failure, and all-cause mortality. While following almost 9000 individuals, an increase in hs-cTnT was independently associated with incident CHD, death, and heart failure.[8] In parallel, Hughes et al added important information about the predictive value of repeated measurements of hs-cTnI. In their study they took 3 troponin samples 5 years apart from each other in 3875 individuals initially from CHD and found the following patterns: hs-cTnI increased over time and this increase was associated with an increase in fatal and non-fatal CVD events. However, the repeat measures of hs-cTnI obtained at baseline, 5, and, 10 years did not significantly improve risk prediction compared with a single measurement in this primary care setting.[7] In patients with chronic heart failure, changes in hs-cTnT (either at 3 or 4 months after the initial visit) were also associated with future CVD events, but only marginally improved the net prediction generated by a single measurement.[16] In 2285 patients suffering from type 2 diabetes mellitus and stable CHD, Everett et al found troponin T (measured by a conventional assay) to independently associate with cardiovascular mortality, myocardial infarction, or stroke.[17]

Table 4. Cox-Proportional Hazard Models for Association of hs-cTnT With Fatal and Non-Fatal CVD Events Stratified According to Presence of Myocardial Infarction at Baseline (Versus Coronary Heart Disease)

	Models Adjusted for Age and Sex		Fully Adjusted Model*		c-Statistic	
hs-cTNT	HR (95% CI)	*P* Value	HR (95% CI)	*P* Value	AUC (95% CI)	NRI
Patients with myocardial infarction at baseline						
Basic model*					0.69 (0.63–0.74)	
Baseline level of hs-cTnT	1.29 (1.00–1.66)	0.049	1.09 (0.82–1.46)	0.56	0.69 (0.64–0.74)	0.005 (*P*=0.76)
12-month level of hs-cTnT	1.34 (1.00–1.79)	0.051	1.15 (0.84–1.57)	0.38	0.69 (0.64–0.74)	0.023 (*P*=0.39)
Simultaneously adjusted						
Baseline level of hs-cTnT	1.20 (0.91–1.59)	0.20	1.05 (0.77–1.43)	0.76		
12-month level of hs-cTnT	1.23 (0.89–1.69)	0.21	1.13 (0.81–1.58)	0.48	0.69 (0.64–0.74)	0.009 (*P*=0.75)
Patients without myocardial infarction						
Basic model*					0.71 (0.66–0.76)	
Baseline level of hs-cTnT	2.80 (2.03–3.88)	<0.0001	2.33 (1.59–3.42)	<0.0001	0.74 (0.68–0.79)	0.164 (*P*<0.0001)
12-month level of hs-cTnT	3.47 (2.43–4.97)	<0.0001	2.61 (1.70–4.00)	<0.0001	0.74 (0.68–0.79)	0.118 (*P*=0.005)
Simultaneously adjusted						
Baseline level of hs-cTnT	1.74 (1.15–2.64)	0.0085	1.71 (1.08–2.70)	0.021		
12-month level of hs-cTnT	2.33 (1.49–3.65)	0.0002	1.91 (1.17–3.11)	0.0094	0.74 (0.69–0.80)	0.164 (*P*=0.0003)

AUC indicates area under the curve; CVD, cardiovascular disease; HR, hazard ratio; hs-cTnT, high sensitivity cardiac troponin T; NRI, net reclassification improvement.
*Adjusted for age, sex, bone mass index, systolic blood pressure, left ventricular function, smoking status, diabetes mellitus, total cholesterol, high-density lipoprotein cholesterol, CKD Epi (cystatin C-based) and use of statins.

Reports about repeat measurements of hs-troponins in patients with stable CHD to further improve risk stratification are scarce. Bonaca et al addressed this issue by obtaining hs-cTnI values at baseline and 4 months in a total of 3209 patients who survived an acute coronary syndrome. As already seen in individuals free from disease at baseline, hs-cTnI was associated with an increased risk of fatal CVD events along the 2 measurements. Moreover, patients with high hs-cTnI at 30 days (>9 ng/L) were reported to profit from an intensive statin therapy (atorvastatin 80 mg/day) compared with moderate therapy (pravastatin 40 mg/day) in terms of future cardiovascular events.[9]

The complex interplay of troponin levels, statin therapy, and recurrent cardiovascular events was further elucidated by a recent report from Ford et al.[6] 3318 individuals with elevated LDL-cholesterol levels were randomly assigned to receive either pravastatin 40 mg/day or placebo over 5 years within the West of Scotland Coronary Prevention Study (WOSCOPS). Hs-cTnI levels were obtained at baseline and at 1 year. The main findings were first, baseline troponin and change at 1 year were independently associated with future cardiovascular events such as myocardial infarction or death from CHD, and second, pravastatin significantly reduced troponin levels over time irrespectively to its impact on lowering LDL cholesterol. These observations arise the question to which extent hs-troponins might have the potential to mirror the extent of traditional risk factors and/or the success of therapeutic interventions.

In our cohort, the absence of hypertension went along with a more stringent reduction of both troponin levels at 12-months. This finding supports the observation within a recent trial of 8571 patients free of CVD, in which persistent elevated blood pressure associated independently with graded increases of hs-cTnT over 6 years of follow-up.[18] In addition, even the history of diabetes mellitus was associated with a smaller reduction of both hs-troponins compared with individuals free from hyperglycemia in our analyses. The observation that diabetes mellitus (expressed by HbA_1c) also results in elevated hs-cTnT levels has been made before.[19] Rubin et al showed within almost 10 000 patients free from CHD that there is a linear relation between HbA_1c levels and hs-cTnT suggesting subclinical myocardial injury driven by hyperglycemia. Also, McEvoy et al observed within the ARIC (Atherosclerosis Risk in Communities Cohort) Study an association between consistently elevated fasting glucose levels and elevated hs-cTnT over 6 years of follow-up.[18] Likewise, an elevated BMI was associated with elevated levels of hs-cTnT in ARIC.[18] The latter important finding has been substantiated within our cohort as an elevated BMI not only associates with an absolute increase of hs-cTnT at 1-year follow-up, but also with an increase of hs-cTnI. However, whether an intensive control of such traditional risk factors

Table 5. Cox-Proportional Hazard Models for Association of hs-cTnI With Fatal and Non-Fatal CVD-Events Stratified According to Presence of Myocardial Infarction at Baseline (Versus Coronary Heart Disease)

	Models Adjusted for Age and Sex		Fully Adjusted Model*		c-Statistic AUC (95% CI)	NRI
	HR (95% CI)	*P* Value	HR (95% CI)	*P* Value		
Patients with myocardial infarction at baseline						
Basic model*					0.69 (0.63–0.74)	
Baseline level of hs-cTnI	1.09 (0.83–1.42)	0.55	0.94 (0.68–1.29)	0.69	0.69 (0.63–0.74)	0.025 (*P*=0.068)
12-month level of hs-cTnI	1.30 (1.06–1.60)	0.011	1.17 (0.94–1.47)	0.17	0.69 (0.64–0.75)	0.035 (*P*=0.11)
Simultaneously adjusted						
Baseline level of hs-cTnI	0.96 (0.71–1.30)	0.79	0.86 (0.61–1.21)	0.39		
12-month level of hs-cTnI	1.32 (1.06–1.64)	0.014	1.21 (0.96–1.53)	0.10	0.69 (0.64–0.75)	0.018 (*P*=0.35)
Patients without myocardial infarction						
Basic model*					0.71 (0.66–0.76)	
Baseline level of hs-cTnI	1.79 (1.52–2.12)	<0.0001	1.59 (1.30–1.95)	<0.0001	0.75 (0.71–0.80)	0.137 (*P*<0.0001)
12-month level of hs-cTnI	2.23 (1.81–2.74)	<0.0001	1.87 (1.45–2.40)	<0.0001	0.76 (0.71–0.81)	0.110 (*P*=0.0025)
Simultaneously adjusted						
Baseline level of hs-cTnI	1.20 (0.89–1.62)	0.23	1.22 (0.88–1.68)	0.24		
12-month level of hs-cTnI	1.86 (1.30–2.66)	0.0008	1.55 (1.05–2.30)	0.029	0.76 (0.71–0.81)	0.150 (*P*<0.0001)

AUC indicates area under the curve; CVD, cardiovascular disease; HR, hazard ratio; hs-cTnT, high sensitivity cardiac troponin T; NRI, net reclassification improvement.
*Adjusted for age, sex, bone mass index, systolic blood pressure, left ventricular function, smoking status, diabetes mellitus, total cholesterol, high-density lipoprotein cholesterol, CKD Epi (cystatin C-based) and use of statins.

actually translates into a detectable decrease of subclinical myocardial injury and subsequently lower hs troponins over time, remains obscure at this time.[18]

Beside the predictive value of repeat measurements of hs troponins, also NT-proBNP as an additional well-established cardiovascular biomarker of left ventricular function proved to be even stronger and independently associated with recurrent cardiovascular events at 12 months compared with baseline levels.[12] Hence, these biomarkers tend to not only to be helpful in the acute setting, but also have the potential to classify and discriminate patients being at high risk for recurrent CVD events when used through follow-up visits.

Strength and Limitations

A strength of our analyses is the long-term follow-up over 12 years and a high response rate during follow-up. Also, the comprehensive characterization of our cohort (including several traditional and novel risk factors) allowed us to generate fully adjusted models. Despite this reasonable conceptual framework, some limitations have to be taken into account while interpreting our results. First, our analyses are based on 2 troponin samples at baseline and after 12 months of follow-up rather than several repeat measurements to address day-to-day variability. Although troponin concentrations might even show circadian fluctuations,[20] our approach is in line with previous reports about repeat measurements of biomarkers. Moreover, the short- and long-term individual variation in patients with symptoms of stable CHD appears to be mild.[15] Second, critically ill patients after the initial event might not have been able to take part in a 3-week rehabilitation program and were therefore potentially underrepresented in our analyses. However, given that our results were based on a slightly less severe ill population, the actual predictive effect should be even stronger in more seriously affected patients. Third, unfortunately subgroup analyses (eg, stratified by sex or type of secondary cardiovascular event) are unrewarding given the limited sample size of our cohort. Fourth, data about the quality of risk factor management (eg, control of hypertension, diabetes mellitus) are suboptimal. Hence, the hs-troponin levels obtained within our study are not suitable to mirror the actual quality of secondary prevention in detail. Fifth, all troponin samples underwent one freeze-thaw cycle before analysis. Although there is evidence showing that even 2 freeze-thaw cycles do not substantially affect the sample stability,[21] data on the long-term stability of such samples at −80°C are not available to date.[22] Sixth, isoforms of both hs-troponins are expressed in the myocardium as well as in the skeletal muscle. However, the cross-reactivity of hs-cTnI assays might be lower compared with hs-cTnT assays. Hence, the cardiac specificity of these assays might differ and as a result the prognostic value, too.[23] Seventh, since 85% of the

Table 6. Relative Change of Log-Transformed hs-cTnT and Log-Transformed hs-cTnI by Categories and as Continuous Variable, Cox-Proportional Hazard Models

Category (min, max)	Events/Subjects	Incidence Rate (Per 1000 person-years)	Log-Rank Test	Adjusted for Log-Transformed Baseline Levels of hs-cTnT or hs-cTnI, Age and Sex		Fully Adjusted Model*	
				HR (95% CI)	*P* Value	HR (95% CI)	*P* Value
hs-cTnT, ng/L							
1 (−88.79 to −30.76)	15/111	15.0	0.20	Reference			
2 (−30.72 to −16.44)	29/114	27.2		1.90 (1.01; 3.58)	0.047	1.73 (0.90; 3.32)	0.10
3 (−16.43 to 0)	33/134	27.8		2.56 (1.37; 4.79)	0.0033	2.03 (1.06; 3.92)	0.034
4 (0.25 to 660.09)	14/70	22.4		4.17 (1.90; 9.14)	0.0004	3.01 (1.35; 6.73)	0.0072
Relative change as a continuous variable				1.007 (1.002; 1.012)	0.0040	1.006 (0.999; 1.012)	0.072
hs-cTnI, ng/L							
1 (−80.36 to −31.71)	17/95	19.8	0.36	Reference			
2 (−31.70 to −17.55)	19/107	18.8		0.87 (0.45; 1.69)	0.69	0.68 (0.33; 1.40)	0.29
3 (−17.32 to 0)	29/129	25.8		1.58 (0.86; 2.90)	0.14	1.25 (0.66; 2.36)	0.49
4 (0.12 to 184.99)	26/98	29.5		2.44 (1.29; 4.62)	0.0062	1.73 (0.88; 3.37)	0.11
Relative change as a continuous variable				1.014 (1.005; 1.024)	0.0028	1.009 (0.999; 1.020)	0.079
Simultaneously adjusted							
hs-cTnT, ng/L							
1 (−88.79 to −30.76)				Reference			
2 (−30.72 to −16.44)				1.75 (0.88; 3.49)	0.11	1.63 (0.82; 3.25)	0.17
3 (−16.43 to 0)				2.38 (1.17; 4.84)	0.017	1.81 (0.87; 3.76)	0.11
4 (0.25 to 660.09)				3.07 (1.29; 7.30)	0.011	2.53 (1.06; 6.07)	0.037
Relative change as a continuous variable				1.004 (0.998; 1.010)	0.22	1.003 (0.996; 1.011)	0.38
hs-cTnI, ng/L							
1 (−80.36 to −31.71)				Reference			
2 (−31.70 to −17.55)				0.68 (0.33; 1.38)	0.28	0.60 (0.29; 1.25)	0.17
3 (−17.32 to 0)				1.13 (0.58; 2.19)	0.72	0.99 (0.51; 1.94)	0.98
4 (0.12 to 184.99)				1.69 (0.83; 3.44)	0.15	1.44 (0.69; 3.03)	0.33
Relative change as a continuous variable				1.013 (1.003; 1.022)	0.0090	1.009 (0.998; 1.020)	0.12

Patients without myocardial infarction at baseline. HR indicates hazard ratio

*Adjusted for log-transformed baseline value of hs-cTnT or hs-cTnI, age, sex, bone mass index, history of high blood pressure, left ventricular function, smoking status, diabetes mellitus, total cholesterol, high-density lipoprotein cholesterol, CKD Epi (cystatin C-based) and use of statins.

study participants were male, the applicability of the results for a more balanced cohort is disputable.

Conclusions

In our cohort consisting of almost 900 patients with stable CHD, hs-troponins obtained during 12-months follow-up after an inpatient rehabilitation program following an acute cardiovascular event were associated even stronger with recurrent events than the baseline values in patients with CHD and without a baseline MI, a finding, which needs further corroboration. In a model controlling for several traditional and rather novel risk factors, the longitudinal changes of such biomarkers seem to improve the risk prediction beyond single measurements taken shortly after the initial cardiovascular event. Thus, the evaluation of repeat troponin measurements during follow-up in this patient group might help to identify those patients being at high risk for recurrent events emphasizing the vigorous control of risk factors in such patients.

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