Prognostic implications of high-sensitivity cardiac troponin T assay in a real-world population with non-ST-elevation acute coronary syndrome

Marco Magnoni a,⁎, Guglielmo Gallone a, Ferruccio Ceriotti a, Vittoria Vergani a, Daniela Giorgio a, Giulia Angeloni b, Attilio Maseri c, Domenico Cianflone a

a IRCCS Ospedale San Raffaele, Università Vita-Salute San Raffaele, Milan, Italy
b Department of Heart and Vessels, Careggi Hospital, University of Florence, Florence, Italy
c Heart Care Foundation Onlus, Florence, Italy

1. Introduction

High-sensitivity troponin assays (hs-cTn) are able to detect circulating cardiac troponin (cTn) levels at an order of magnitude lower than contemporary ones. HS-cTn were proven to provide earlier detection of acute myocardial infarction (MI) [1,2] compared to contemporary ones. Hs-cTn were proven to provide earlier detection of acute myocardial infarction (MI) [1,2] compared to contemporary ones. HsTnT may provide a more accurate prognostic stratification than contemporary cardiac troponin I (cTnI) in patients with non-ST-segment elevation acute coronary syndromes (NSTE-ACS).

Methods: HsTnT and cTnI were measured in 644 patients with CK-MB negative NSTE-ACS who were enrolled in the prospective multicenter SPAI (Stratificazione Prognostica dell’Angina Instabile) study. Patients were stratified at the 99th percentile reference limit for each assay. The primary endpoint was cardiovascular death (CVD) or non-fatal myocardial infarction (MI); the secondary endpoint was the occurrence of unstable angina (UA). Follow-up lasted 180 days.

Results: Patients with hsTnT ≥99th percentile were at higher risk of CVD/MI (30-day: 5.9% vs 0.8%, p = 0.001; 180-day: 11.1% vs 4.7%, p = 0.004), also after adjusting for TIMI Risk Score. No significant difference in CVD/MI at 180-day was found between hsTnT-positive/cTnI-negative and hsTnT-negative/cTnI-negative patients (adjHR 1.61, 95% CI 0.74–3.49, p = 0.232). Occurrence of UA was not differently distributed between hsTnT groups dichotomized at the 99th percentile (12.4% vs 12.5%, p = 0.54).

Conclusions: Our investigation on a real-world NSTE-ACS population showed good prognostic performance of hsTnT in the risk stratification of the hard endpoint, but did not demonstrate the improved prognostic ability of hsTnT over contemporary cTn. Neither troponin assay predicted the recurrence of UA, suggesting the acute rise of cardiac troponin as a marker of severity, but not the occurrence of future coronary instability.
2. Methods

2.1. Study design and data collection

The population of the current analysis was enrolled in the SPAI study (Stratificazione Prognostica dell’Angina Instabile), a prospective, observational, multi-centric Italian study designed to investigate the prognostic value of clinical variables and circulating biomarkers in patients admitted to a coronary care unit (CCU) with a diagnosis of UA according to CK-MB negativity. Details about the SPAI study are described elsewhere [16]. The costs were supported by Regione Calabria and by Fondazione per il Cuore ONLUS, and coordination by Centro Ricerche Coronariche at Università Cattolica del Sacro Cuore.

A total of 983 consecutive patients with suspected UA and admitted to the CCU of 21 Italian Cardiology Units (7 including CCU exclusively, 14 with a cath lab of which 12 with a Cardiac Surgery Unit) from 1997 to December 2001 were enrolled in the study. UA was defined as new onset angina (<2 months), occurring with minimal exertion or at rest (de novo angina) or worsening angina presenting in patients with either an old myocardial infarction or a known history of stable coronary artery disease (destabilizing angina).

Inclusion criteria were: diagnosis of UA confirmed by one or more among a) ischemic ECG changes during recurrent chest pain; b) evidence of myocardial ischemia during exercise ECG stress test or exercise radionuclide studies or pharmacological (dipyridamole or dobutamine) echocardiographic stress tests; c) documentation of obstructive (>50%) stenosis in at least one major epicardial artery during coronary angiography.

Exclusion criteria were: a) failure to confirm UA, b) diagnosis of MI, as defined by the World Health Organization [17], documented by 12 lead ECG monitoring and CK-MB blood levels at admission and after 6, 12 and 24 h, c) hospital readmission within 3 months after discharge for an acute myocardial infarction because of recurrent angina (post-MI UA), d) evidence of reduced left ventricular systolic function (EF ≥40%) on two-dimensional echocardiography, e) severe infectious comorbidities.

Clinical data, standard ECG, and venous blood samples were obtained upon admission. Patients were treated according to local standard protocols.

All subjects provided written informed consent to take part in the study. The study was carried out in accordance with the Declaration of Helsinki and was approved by the institutional ethics committee.

2.2. Study population

983 patients with suspected UA were admitted in the 21 CCU participating in the SPAI study. The diagnosis was confirmed in 845 patients. 46 patients with post-MI UA and 8 patients with infectious co-morbidities were excluded. HsTnT was not analyzed for 147 patients because of frozen serum samples were no more available. Thus, 644 patients were included in our study.

2.3. Troponin testing

Baseline blood samples were obtained at admission. Blood was centrifuged, and serum and plasma were stored at −80 °C. All circulating biomarkers were measured in a central laboratory, in a single batch, by personnel unaware of patients’ characteristics. Plasma cTnI was measured with an immunoassay system (Stratus Dade Troponin I assay, Behring), which has a limit of detection (LoD) of 0.1 ng/mL and a 99th percentile reference limit of 0.4 ng/mL. Plasma hsTnT was measured with a high-sensitivity assay (Elecsys Troponin T high-sensitive assay, Roche Diagnostics), which has a LoD of 5 ng/L, a 99th percentile reference limit of 14 ng/L and a coefficient of variation of 10% at 13 ng/L [18].

2.4. Outcome measures

Patients were followed-up at 90 and 180 days after discharge by out-patient clinical visits or by telephone interview. In case of death, detailed information was obtained from clinical records or the patient’s physician or relatives. Death was considered of cardiac origin when it was consequent to MI or heart failure, or when sudden.

The primary endpoint was a composite of CVD and non-fatal MI during hospitalization and the 180-day follow-up. MI was diagnosed in case of chest pain lasting >30 min with ischemic ECG changes and a rise in CK-MB. The secondary endpoint was the future occurrence of UA requiring hospitalization during the 180-day follow-up.

2.5. Statistical analyses

The distribution of baseline characteristics for categorical variables is expressed as percentage frequency. Continuous variables are described as means and standard deviations or as medians and interquartile ranges, as appropriate. Differences among groups were detected using Pearson χ² test and Fisher’s exact test for categorical variables and Mann-Whitney test for continuous variables.

Table 1 Baseline characteristics by hs-TnT levels.

| Groups | hsTnT < 14 ng/L | hsTnT ≥ 14 ng/L | p value |
|--------|----------------|----------------|---------|
| n = 240 |                 |                |         |
| General characteristics |
| Age (years) | 64 (57–70) | 69 (60–75) | <0.0001 |
| Male (%) | 57.7 | 69.1 | 0.002 |
| Medical history |
| Family history (%) | 34.7 | 39.6 | 0.125 |
| Hypertension (%) | 90.4 | 88.5 | 0.370 |
| Hypertension (%) | 59.2 | 54.8 | 0.158 |
| Diabetes (%) | 20.4 | 26.7 | 0.043 |
| Current smokers (%) | 24.8 | 28.6 | 0.169 |
| BMI | 26 (24–28) | 26 (24–28) | 0.886 |
| Previous CAD history |
| Chronic angina (%) | 21.2 | 24.5 | 0.198 |
| AMI (%) | 18.8 | 25.0 | 0.040 |
| UA (%) | 14.6 | 21.7 | 0.015 |
| PCI (%) | 8.8 | 6.9 | 0.244 |
| CABG (%) | 5.2 | 8.3 | 0.081 |
| Presentation |
| Previous 48 h instability episodes | |
| 0 | 6.7 | 6.9 | 0.423 |
| 1–2 | 64.7 | 60.5 |
| >3 | 28.6 | 32.5 |
| cTnI (ng/mL) | 1.0 (0.04–1075) | 0.42 (0.14–1075) | <0.0001 |
| HsTnT (ng/L) | 6.90 (4.23–9.88) | 8.77 (29.61–161.15) | <0.0001 |
| CRP (mg/L) | 3.15 (1.50–6.93) | 5.73 (2.56–16.04) | <0.0001 |
| TIMI 2–3 (%) | 66.7 | 75.5 | 0.010 |
| ST segment depression | |
| 60.4 | 81.7 | <0.0001 |
| GFR | 73 (63–84) | 68 (54–83) | 0.001 |

Treatment after enrolment |
| ASA (%) | 91.7 | 93.6 | 0.226 |
| DAPT (%) | 25.0 | 20.0 | 0.086 |
| α β-blockers (%) | 61.3 | 53.5 | 0.032 |
| ACE-I/ARB (%) | 31.2 | 23.3 | 0.017 |
| Statin (%) | 34.6 | 26.2 | 0.016 |
| DHP-CBB (%) | 32.5 | 31.2 | 0.397 |
| NDHP-CBB (%) | 33.8 | 32.3 | 0.372 |
| Heparin/LMWH (%) | 46.2 | 43.6 | 0.280 |
| PCI (%) | 15.4 | 17.6 | 0.276 |
| CABG (%) | 2.5 | 3.7 | 0.276 |

Abbreviations: HsTnT, high-sensitivity cardiac troponin T; BMI, body mass index; CAD, coronary artery disease; AMI, acute myocardial infarction; UA, unstable angina; PCI, percutaneous coronary intervention; CABG, coronary artery bypass graft; cTnI, conventional cardiac troponin I; CRP, C-reactive protein; TIMI, thrombolysis in myocardial infarction; GFR, glomerular filtration rate; DAPT, dual antiplatelet therapy; ACE-I, angiotensin-converting enzyme inhibitor; ARB, angiotensin receptor blocker; DHP-CBB, dihydropiridiné calcium-channel blocker; NDHP-CBB, non dihydropiridiné calcium-channel blocker; LMWH, low molecular weight heparin
Event rates at 30 and 180 days were stratified by guideline-based cTnI and hsTnI 99th percentile reference limits (0.4 ng/mL and 14 ng/L respectively). Cox regression analysis was used to evaluate the relation between hsTnT and outcome. Adjusted analyses took into account all the remaining elements of the TIMI risk score [19] including age, recent aspirin use, ≥3 CV risk factors, known coronary disease, ST segment deviation and repeated episodes of rest angina in the last 24 h. The differences in endpoints were analyzed with the log-rank test and expressed as Kaplan Meier curves. A p value<0.05 was considered statistically significant.

All statistical analyses were performed with JMP statistical software (version 11.0.0, SAS Institute Inc., Cary, North Carolina, USA) and SPSS Statistics (version 20, IBM Corporation, Armonk, NY, USA).

3. Results

3.1. Baseline characteristics

Baseline characteristics are presented and dichotomized at the hsTnT 99th percentile reference limit of 14 ng/L in Table 1. Significant differences were observed between the two groups of patients. Patients with hsTnT ≥14 ng/L were prevalently males and older, more frequently had a known history of diabetes, myocardial infarction, and UA, and more commonly presented with higher TIMI risk score, depressed ST segment, lower glomerular filtration rate (GFR) and higher levels of C-reactive protein (CRP). ACE-inhibitors/ARBs and statins were more commonly prescribed in patients with hsTnT <14 ng/L and no additional significant differences in treatment were seen.

3.2. Diagnostic performance of hsTnT

cTnI assays revealed the presence of myocardial necrosis in 215 (33.4%) patients, whereas hsTnT was detected above the 99th hsTnT percentile reference limit in 404 (62.7%) patients, thus providing a reclassification ability from UA to NSTEMI of 29.3% over cTnI (Fig. 1).

3.3. Prognostic performance of hsTnT: primary endpoint

Over the 180-day follow-up, 56 (8.7%) patients had an MI or died of cardiac death (Table 2).

3.3.1. HsTnT quartiles

A significant graded increase in primary endpoint occurrence was observed across hsTnT quartiles at 180 days (3.7%, 6.2%, 11.7%, 13.2%; p-trend = 0.002) and was already present at 30 days (0.6%, 1.9%, 6.1%, 7.5% p-trend = 0.003) (Fig. 2).

3.3.2. HsTnT 99th percentile reference limit

In order to define the prognostic ability of hsTnT according to current international guidelines, we analyzed the population according to the 99th percentile reference limit (Fig. 2). At 30 and 180 days, patients with hsTnT ≥14 ng/L had a significantly higher rate of CVD or MI (30-day: 5.9% vs 0.8% p = 0.001; 180-day: 11.1% vs 4.7% p = 0.002). When adjusting for the remaining elements of TIMI risk score, patients with hsTnT ≥14 ng/L had a 2.24-fold higher risk of CVD or MI (30-day: adj HR 6.35, 95% CI 1.49–27.01, p < 0.001; 180-day adj HR 2.24, 95% CI 1.15–4.35, p < 0.001).

3.3.3. Comparison with the cTnI assay

To investigate the added value of the high sensitivity assay compared to the former one, the study population was divided into four groups according to hsTnT and cTnI 99th percentile reference limits as follows: UA (hsTnT-negative), n = 233; Reclassified NSTEMI (hsTnT-positive, cTnI-negative), n = 196; Traditional NSTEMI (hsTnT-positive, cTnI-positive), n = 208; group 4 (hsTnT-negative, cTnI-positive), n = 7. No primary endpoint event was recorded in group 4, similarly to previously reported findings [13]. A significant gradient of risk for CVD/MI across the remaining three groups was present at 30 days (0.9%, 3.1%, 8.7%; p-trend<0.001) and persisted at 180 days (4.7%, 8.2%, 13.9%; log-rank = 0.004) (Table 2). At the direct comparison, the “Reclassified NSTEMI” group showed higher adverse event rates than the “UA” groups, but the differences were not statistically significant either at 30 days (adj HR 3.59, 95% CI 0.72–17.77, p = 0.118) or at 180 days (adj HR 1.61, 95% CI 0.74–3.49, p = 0.232) (Fig. 3).

3.4. Secondary endpoint

At 180-day follow-up, 80 patients (12.4%) had a UA event. No significant difference in UA occurrence was observed across hsTnT quartiles (14.9%, 9.4%, 15.4%, 10.3% p-trend = 0.234) and between 99th percentile-dichotomized hsTnT groups (12.4% vs 12.5% p = 0.548).
directly comparing the hsTnT and cTnI assays, no differences in secondary endpoint rates were observed among UA, Reclassified NSTEMI, Traditional NSTEMI groups (12.9%, 11.8%, 13.2%; log-rank = 0.973) (Fig. 3).

Of note, we studied the association between hsTnT and 180-day acute coronary events as a whole (i.e., UA and MI), but, as for predictability of UA events, no significant association was observed (hsTnT 99th cut-off: 16.8% vs. 14.6% p = 0.262).

4. Discussion

In the current study, we analyzed and compared the performances of a hsTnT assay and a cTnI assay in a CK-MB negative NSTE-ACS population, in relation to different end-points.

Our study confirms the well-established superior diagnostic accuracy of hs-cTn [1–3] to identify myocardial infarction. We also observed a robust prognostic performance at the guidelines-based 99th percentile reference limit, which identified patients with a 6.4 and a 2.4-fold higher risk of CVD or MI at 30 and 180 days respectively; a significant gradient of risk for adverse outcomes at increasing quartiles of hsTnT concentration was present at the same follow-up intervals. However, the direct comparison of cTnI and hsTnT, despite showing a trend, did not reach statistical significance in demonstrating the hypothesized improved prognostic ability of hs-cTnT over cTn.

Finally, we showed that neither troponin assay predicted the occurrence of UA at 180 days.

Although a broad body of knowledge on diagnostic and prognostic abilities of hs-cTn assays has been developed in the recent years, the clinical relevance of these findings continues to be debated and requires further characterization.

Our study differs in important features from previous works investigating hs-cTn assays and therefore may provide new insight into the clinical impact of this technology.

First, NSTE-ACS populations of previous studies were mainly enrolled in clinical trials with specific, mostly high-risk, inclusion criteria, and were managed according to precise treatment protocols in highly specialized healthcare structures [10, 12–14]. The SPAI population, instead, was enrolled in a prospective observational study involving 21 Italian Cardiology centers with different treatment capabilities, and patients were managed according to local protocols. Therefore, our population is more representative of the real-life general NSTE-ACS population. Moreover, the SPAI population includes only CK-MB negative patients; as such, it exclusively investigates the range of NSTE-ACS spectrum where the diagnostic and prognostic accuracy of a more sensitive assay has a greater clinical impact.

Second, we chose a cTnI assay as a benchmark, while all previous studies except one [13] used a cTnT assay. Notably, we found a very low proportion of cTnI-positive patients in the hsTnT-negative group (2.9%), similarly to what was observed in the other study that used cTnI as comparator (4.2%) [13], and in contrast to the very high proportion of cTnT-positive patients in the hs-cTn-negative group reported in the studies that instead used cTnT as benchmark (81.6% and 77.1% in Bohula and Bonaca studies using cTnI as comparator (2.9 vs 9.2% [12], 1.9% vs 8.2% [14] versus 4.7% vs 8.2% [our study], 3.8% vs 7.0% [13]). Accordingly, we believe the poor precision of the fourth-generation cTnT assay at the low end of concentration, rather than a class effect of hs-cTn over fourth-generation assays. Indeed, in the latter studies, the magnitude of difference in hard endpoints between the cTn neg/hs-cTn neg group and the cTn neg/hs-cTn pos group was considerably greater than in our and Bonaca studies using cTnI as comparator (2.9 vs 9.2% [12], 1.9% vs 8.2% [14] versus 4.7% vs 8.2% [our study], 3.8% vs 7.0% [13]). Accordingly, we believe the poor precision of the fourth-generation cTnT assay at the low end of concentration to contribute at least partially to the gain in hs-cTnT prognostic ability over cTnT demonstrated in those studies, which may thus be overestimated.

Even if the analysis of a larger group of patients - in consideration of the positive trend observed - would have probably led to a statistically significant result as observed in Bonaca et al. [13], our observations at least partially downsize the clinical significance of previous reports. These findings highlight once again how the wide variability in assays analytical features do not allow generalization of their diagnostic and prognostic value without direct clinical evaluation of each assay [20, 21].

Last, we analyzed the prognostic ability of hs-cTn also concerning future UA following NSTE-ACS, whereas to date studies only focus on hard endpoints, i.e., CVD and MI. The importance to include a broad prognostic characterization, also encompassing the recurrence of UA, in the validation process of hs-cTnT assays for clinical practice use has been vigorously stressed by expert opinion [15]. Our analysis found no association of either conventional or high-sensitivity assay with UA occurrence. This observation may suggest that troponin acute raise acts as a marker of severity of the potential subsequent event, rather than predicting the probability of event recurrence, since it stratifies risk of CVD/MI, but not of UA.

| Table 2 | CVD/MI and UA rates at 30 and 180 days according to cTnT/hsTnT classes. |
|----------------|-----------------|-----------------|-----------------|-----------------|
|                 | n = 233         | n = 196         | n = 208         | n = 7           |
| **Death/AMI**   |                 |                 |                 |                 |
| 30 days         | 0.9% Reference group | 3.1% adj HR 3.59 (0.72–17.77) p = 0.118 | 8.7% adj HR 9.09 (2.08–40.00) p = 0.003 | 0.0% | -0.0001 |
| 180 days        | 4.7% Reference group | 8.2% adj HR 1.61 (0.74–3.49) p = 0.232 | 13.9% adj HR 2.64 (1.30–5.35) p = 0.007 | 0.0% | 0.02 |
| **Unstable angina** |                 |                 |                 |                 |
| 30 days         | 3.5%            | 3.1%            | 4.5%            | 0.0%            | 0.707 |
| 180 days        | 12.9%           | 11.8%           | 13.2%           | 0.0%            | 0.875 |

Abbreviations: Adj HR, adjusted hazard ratio; hsTnT, high-sensitivity cardiac troponin T; cTnI, conventional cardiac troponin I; AMI, acute myocardial infarction; UA, unstable angina; NSTEMI, non-ST segment elevation myocardial infarction.
These results are consistent with the concept that different biomarkers reflect different features of the pathobiology in NSTE-ACS, thus predicting distinct aspects of the disease clinical spectrum [22, 23]. Our study has several limitations mainly related to its observational nature. Patients from the SPAI population were treated according to hospital local protocols, as such only a minority of patients (21%) underwent revascularization procedures. More recent clinical trials and current clinical practice have higher rates of early invasive treatment. However, recent evidence suggests that a conservative treatment strategy may be appropriate in hs-cTn-negative patients [24]. Accordingly, our study results may be generalizable to a wide proportion of low-risk NSTE-ACS patients. Finally, as only blood samples at presentation were available, we could not evaluate the prognostic implications of the “delta -approach” showed in other studies [25].

In conclusion, the hsTnT assay evaluated in this study demonstrated to have a good prognostic performance in the risk stratification of hard endpoint risk stratification, but it did not show the improved prognostic ability of hsTnT over cTn reported in previous studies.

Neither troponin assay predicted the occurrence of UA, suggesting cTn acute raise to be a marker of severity rather than recurrence probability of the subsequent event. Hence, the importance of the independent predictive value of different factors and the need to differentiate end point in order to carry out and improve identification of subgroups at risk.

Conflict of interest

The authors report no relationships that could be construed as a conflict of interest.

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References

[1] S.J. Aldous, M. Richards, L. Cullen, R. Troughton, M. Than, Diagnostic and prognostic utility of early measurement with high-sensitivity troponin T assay in patients presenting with chest pain, CMAJ 184 (5) (2012) 260–268.
[2] N.L. Mills, Implementation of a sensitive troponin I assay and risk of recurrent myocardial infarction and death in patients with suspected acute coronary syndrome, JAMA 305 (12) (2011) 1210.
[3] E. Giannitsis, M. Becker, K. Kurz, G. Hess, D. Zdunek, H.A. Katus, High-sensitivity cardiac troponin T for early prediction of evolving non-ST-segment elevation myocardial infarction in patients with suspected acute coronary syndrome and negative troponin results on admission, Clin. Chem. 56 (4) (2010) 642–650.
[4] E.W. Carlton, L. Cullen, M. Than, J. Gamble, A. Khattab, K. Greaves, A novel diagnostic protocol to identify patients suitable for discharge after a single high-sensitivity troponin, Heart 101 (13) (2015) 1041–1046.
[5] R. Body, S. Carley, G. McDowell, A.S. Jaffe, M. France, K. Cruickshank, et al., Rapid exclusion of acute myocardial infarction in patients with undetectable troponin using a high-sensitivity assay, J. Am. Coll. Cardiol. 58 (13) (2011) 1332–1339.
[6] N. Bandstein, R. Ljung, M. Johansson, M.J. Holmznn, Undetectable high-sensitivity cardiac troponin T level in the emergency department and risk of myocardial infarction, J. Am. Coll. Cardiol. 63 (23) (2014) 2569–2578.
[7] M. Rubini Gímenez, R. Hoeller, T. Reichlin, C. Zellweger, R. Twerenbold, M. Reiter, et al., Rapid rule out of acute myocardial infarction using undetectable levels of cTnI, Int. J. Cardiol. 168 (4) (2013) 3896–3901.
[8] T. Reichlin, W. Hochholzer, S. Basset, S. Steuer, C. Stelzig, S. Hartwig, et al., Early diagnosis of myocardial infarction with sensitive cardiac troponin assays, N. Engl. J. Med. 361 (9) (2009) 858–867.
[9] R. Body, G. Burrows, S. Carley, L. Cullen, M. Than, A.S. Jaffe, et al., High-sensitivity cardiac troponin T concentrations below the limit of detection to exclude acute myocardial infarction: a prospective evaluation, Clin. Chem. 61 (7) (2015) 983–989.
[10] B. Lindahl, P. Venge, S. James, The new high-sensitivity cardiac troponin T assay improves risk assessment in acute coronary syndromes, Am. Heart J. 160 (2) (2010) 224–229.
[11] C. Ndrepepa, S. Braun, S. Schulz, R.A. Byne, J. Pache, J. Meldahl, et al., Comparison of prognostic value of high-sensitivity and conventional troponin T in patients with non-ST-segment elevation acute coronary syndromes, Circ. Res. Acta 412 (15–16) (2011) 1350–1356.
[12] L.A. Bohula May, M.P. Bonaca, P. Jarolim, E.M. Antman, E. Braunwald, R.P. Giugliano, et al., Prognostic performance of a high-sensitivity cardiac troponin I assay in patients with non-ST-elevation acute coronary syndrome, Clin. Chem. 60 (1) (2014) 158–164.
[13] M.P. Bonaca, R.G. O’Malley, S.A. Murphy, P. Jarolim, M.J. Conrad, E. Braunwald, et al., Prognostic performance of a high-sensitivity assay for cardiac troponin I after non-ST elevation acute coronary syndrome: analysis from MERLIN-TIMI 36, Eur Heart J Acute Cardiovasc Care. 4 (5) (2015) 431–440.
[14] L. Grinstein, M.P. Bonaca, P. Jarolim, M.J. Conrad, E. Bohula-May, N. Deeyadavalu, et al., Prognostic implications of low level cardiac troponin elevation using high-sensitivity cardiac troponin T, Clin. Cardiol. 38 (4) (2015) 230–235.
[15] F.S. Apple, High-sensitivity cardiac troponin assays: what analytical and clinical issues need to be addressed before introduction into clinical practice? Clin. Chem. 56 (6) (2010) 886–891.
[16] G.A. Lanza, D. Cianfrone, A.G. Rebuzzi, G. Angeloni, A. Sestito, G. Ciriello, et al., Prognostic value of ventricular arrhythmias and heart rate variability in patients with unstable angina, Heart 92 (8) (2006) 1055–1063.
[17] K. Thygesen, J.S. Alpert, H.D. White, Joint ESC/ACCF/AHA/WHF task force for the re-definition of myocardial infarction. Universal definition of myocardial infarction, Eur. Heart J. 28 (20) (2007 Oct) 2525–2538.
[18] E. Giannitsis, K. Kurz, K. Hallemayer, J. Jarausch, A.S. Jaffe, H.A. Katus, Analytical validation of a high-sensitivity cardiac troponin T assay, Clin. Chem. 56 (2) (2010) 254–261.
[19] E.M. Antman, M. Cohen, P.J. Bernink, C.H. McCabe, T. Horacek, G. Papuchis, et al., The TIMI risk score for unstable angina/non-ST elevation MI: a method for prognostic assessment and therapeutic decision making, JAMA 284 (7) (2000) 835–842.
[20] F.S. Apple, Y. Sandoval, S.W. Smith, F.S. Apple, Present and future of cardiac troponin in clinical practice: a paradigm shift to high-sensitivity assays, Am. J. Med. 129 (4) (2016) 354–365.
[21] F.S. Apple, Y. Sandoval, A.S. Jaffe, J. Ordonez-Llanos, IFCC task force on clinical applications of cardiac biomarkers. Cardiac troponin assays: guide to understanding analytical characteristics and their impact on clinical care, Clin. Chem. 63 (1) (2017) 73–81.
[22] K.M. Eggers, B. Lagerqvist, P. Venge, L. Wallentin, B. Lindahl, Prognostic value of biomarkers during and after non-ST-segment elevation acute coronary syndrome, J. Am. Coll. Cardiol. 54 (4) (2009) 357–364.
[23] C. Mueller, Biomarkers and acute coronary syndromes: an update, Eur. Heart J. 35 (9) (2014) 552–556.

[24] E. Giannitsis, L. Wallentin, S.K. James, M. Bertilsson, A. Siegbahn, R.F. Storey, et al., Outcomes after planned invasive or conservative treatment strategy in patients with non-ST-elevation acute coronary syndrome and a normal value of high sensitivity troponin at randomisation: a platelet inhibition and patient outcomes (PLATO) trial biomarker substudy, Eur Heart J Acute Cardiovasc Care. 6 (6) (2017) 500–510.

[25] T. Reichlin, R. Twerenbold, C. Maushart, M. Reiter, B. Moehring, N. Schaub, et al., Risk stratification in patients with unstable angina using absolute serial changes of 3 high-sensitive troponin assays, Am. Heart J. 165 (3) (2013).