Clinical Chemistry 48:9
1432–1436 (2002)

Enzymes and Protein Markers

Single-Point Cardiac Troponin T at Coronary Care Unit Discharge after Myocardial Infarction Correlates with Infarct Size and Ejection Fraction

Mauro Panteghini,[1*] Claudio Cuccia,[2] Graziella Bonetti,[1] Raffaele Giubbini,[3] Franca Pagani,[1] and Elena Bonini[2]

Background: **One of the major concerns in replacing creatine kinase MB (CK-MB) with cardiac troponins is the lack of evidence of the ability of troponins to estimate the size of acute myocardial infarction (AMI). We investigated the ability of a single measurement of cardiac troponin T (cTnT) at coronary care unit (CCU) discharge to estimate infarct size and assess left ventricular (LV) function in AMI patients.**

Methods: **We studied 65 AMI patients in whom infarct size was estimated by CK-MB peak concentrations and gated single-photon emission computed tomography (SPECT) myocardial perfusion using technetium-99m sestamibi and LV function by SPECT imaging. Measurements of cTnT and SPECT were performed 72 h (median) after admission (range, 40–160 h). SPECT was also repeated 3 months later.**

Results: **We found a significant correlation between cTnT and both the peak CK-MB concentrations ($r = 0.76$; $P < 0.001$) and the perfusion defect size at SPECT ($r = 0.62$; $P < 0.001$). cTnT was inversely related to LV ejection fraction (LVEF) assessed both early ($r = -0.56$; $P < 0.001$) and 3 months after AMI ($r = -0.70$; $P < 0.001$). cTnT >2.98 μg/L predicted a LVEF <40% at 3 months with a sensitivity of 86.7%, specificity of 81.4%, and a likelihood ratio for a positive test of 4.7 (95% confidence interval, 4.0–5.4).**

Conclusions: **A single cTnT measurement at CCU discharge after AMI is useful as a noninvasive estimate of infarct size and for the assessment of LV function in routine clinical setting.**

© 2002 American Association for Clinical Chemistry

The evaluation of infarct size after acute myocardial infarction (AMI)[4] is important for predicting the subsequent clinical course and to validate the effectiveness and clinical relevance of therapeutic interventions *(1–3)*. Quantitative histologic estimates of infarct size are regarded as the gold standard, but the method has little clinical relevance. It is desirable to find a simple and reliable method with which to quantify infarct size *(4, 5)*. Various methods, such as electrocardiography, echocardiography, left ventriculography, radionuclide-based measurements, and the release of cardiac biomarkers, have been proposed *(6)*.

In clinical practice, the extent of injury to the myocardium after AMI is generally assessed by creatine kinase MB isoenzyme (CK-MB) release curves using serial serum sampling *(7, 8)*. Although quantitative calculations based on the area under the CK-MB-vs-time curve are seldom made, many physicians use peak CK-MB to get a qualitative estimate of the size of the infarct *(9, 10)*. The well-known limitations of CK-MB measurements, such as the short duration of increase after AMI, the requirement for repetitive, frequent sampling for evaluation of peak concentrations, the sensitivity to reperfusion status, and the lack of specificity for cardiac damage, have stimulated the search for a more suitable biomarker for sizing infarcts *(11)*.

Cardiac troponin T (cTnT) is a cardiac-specific protein that is compartmented in the contractile apparatus of the myocardial cell. Its release process into the blood after myocardial injury is slow (cTnT is present in plasma for more than 120 h after AMI), and it is only slightly affected

[1] Laboratorio Analisi Chimico Cliniche 1, [2] Divisione di Cardiologia, and [3] Servizio di Medicina Nucleare, Azienda Ospedaliera 'Spedali Civili', 25125 Brescia, Italy.

*Address correspondence to this author at: Laboratorio Analisi Chimico Cliniche 1, Azienda Ospedaliera 'Spedali Civili', Piazza Spedali Civili 1, 25125 Brescia, Italy. Fax 39-030-3995369; e-mail panteghi@bshosp.osp.unibs.it.

Presented in part at the 53rd AACC National Meeting, July 29–August 2, 2001, Chicago, IL (*Clin Chem* 2001;47:A195).

Received March 12, 2002; accepted May 9, 2002.

[4] Nonstandard abbreviations: AMI, acute myocardial infarction; CK-MB, creatine kinase MB isoenzyme; cTnT, cardiac troponin T; CCU, coronary care unit; LV, left ventricular; SPECT, single-photon emission computed tomography; LVEF, LV ejection fraction; and CI, confidence interval.

by reperfusion of the infarct zone *(12)*. For these reasons, plasma cTnT has been used for estimation of infarct size in animals *(13–16)*. In humans, only two clinical studies have examined the correlation of plasma cTnT release to infarct size, and both studies used serial measurements to calculate cTnT peak concentrations and the areas under cTnT curves *(17, 18)*. However, in clinical practice serial analyses are neither feasible nor cost-effective.

In the present study, we investigated the ability of a single-point measurement of circulating cTnT concentrations, performed on the day of discharge from the coronary care unit (CCU), to estimate the infarct size and assess left ventricular (LV) function in AMI patients in comparison with results obtained from gated single-photon emission computed tomography (SPECT) imaging.

Materials and Methods

PATIENT POPULATION

A total of 65 patients with AMI (58 men and 7 women; median age, 53 years; range, 24–78 years) who were admitted to our CCU between May 2000 and June 2001 were prospectively enrolled for this study. These were not consecutive admissions, but no conscious bias was applied in their selection. We included patients who satisfied all the following criteria: (*a*) typical serial changes in serum CK-MB mass concentrations and electrocardiographic evidence of AMI; (*b*) hospitalization within 20 h after the onset of symptoms; (*c*) no evidence of prior AMI; and (*d*) informed consent to allow an extra blood sample to be collected for cTnT analysis at CCU discharge and to undergo SPECT imaging (radiation exposure, 0.6 Rad). The procedures followed were in accordance with the current revision of the Helsinki Declaration. The diagnosis of AMI was made in each patient by a cardiologist without access to cTnT or scintigraphic results. The approximate anatomic region of AMI was determined by electrocardiogram.

Of the 65 patients enrolled, 47 had sustained an anterior and 18 an inferior wall AMI. On admission (median time from symptom onset, 4.5 h; range, 0.5–19 h), all but two patients, who showed left bundle branch block, showed ST elevations or depressions ≥0.1 mV in at least two contiguous leads in their electrocardiograms. Subsequently, new persistent Q waves developed in 39 (60%) patients. Fifty-five (84.6%) patients underwent revascularization: 15 received intravenous thrombolytic therapy, 28 primary percutaneous transluminal coronary angioplasty, and 12 both. The remaining 10 patients were treated with conventional therapy. Except for a patient who died suddenly of a subarachnoid hemorrhage 2 weeks after admission, all patients had an uncomplicated AMI course (no reinfarction) throughout the study period.

PROTOCOL

Serial venous blood (serum) samples for CK-MB peak estimation were taken every 6 h throughout the first 48 h after CCU admission. In addition, for cTnT measurements, a single blood sample was collected into tripotassium EDTA on the morning of the day of CCU discharge (median time after admission, 72 h; range, 40–160 h). Blood samples were centrifuged immediately, and biochemical assays were performed without delay. At the time blood was collected for cTnT, LV function and infarct size were evaluated by gated SPECT imaging. An additional SPECT estimation of LV function was performed in 58 of the enrolled patients ~3 months after hospital discharge. Of the originally enrolled population, one patient died and six patients refused repetition of the SPECT procedure.

LABORATORY ASSAYS

CK-MB mass was measured on an Elecsys® 2010 system (Roche Diagnostics). A previously calculated upper reference limit of 6 μg/L was used. cTnT was measured with the same Elecsys analyzer using the third-generation assay *(19)*. The detection limit of the assay is 0.01 μg/L, and the decision limit used in our hospital for AMI diagnosis is 0.03 μg/L, i.e., the cTnT concentration that meets the imprecision goal of 10% total CV *(20, 21)*. We used CK-MB peak values as biochemical estimates of infarct size *(9, 10)*. A value was defined as a peak if it was the highest in the concentration time course and if we observed at least one lower value before and after this maximum value.

SCINTIGRAPHY

Scintigraphic estimation of infarct size by gated SPECT with technetium-99m sestamibi was performed in resting patients on the day of CCU discharge *(22)*. Quantitative defect size as measurement of infarct size was expressed as the percentage of total LV mass. Gated SPECT imaging also provided information on LV function. In particular, the LV ejection fraction (LVEF) was calculated by dividing the difference between LV end-diastolic volume and LV end-systolic volume by the LV end-diastolic volume. A second SPECT study of LVEF was also performed ~3 months after patients were discharged from the hospital. Procedures and data analysis were performed by independent investigators with no knowledge of patient histories or the results of tests for biochemical markers.

STATISTICAL ANALYSIS

Medians and ranges were calculated to describe continuous variables. Correlations were calculated using standard linear regression analysis. $P < 0.05$ was considered significant. ROC curves were constructed to examine the relationship between cTnT concentration and LVEF. For ROC analysis, the patients were categorized into two datasets, those with a LVEF <40% and those with a LVEF ≥40%, producing a binary classification. The 40% LVEF cutoff had been shown in previous clinical trials to have prognostic significance *(23, 24)*. The ability of cTnT to assign AMI patients accurately into the two groups was determined by calculating the area under the ROC curve. According to the Swets scheme for interpreting the area under the ROC curve *(25)*, values of 0.5–0.7 indicate low diagnostic accuracy, values of 0.7–0.9 suggest limited

clinical utility, and values >0.9 indicate high global diagnostic accuracy. The sensitivity, the specificity, and the likelihood ratio at the best decision value were also calculated. The 95% confidence intervals (CIs) for the population proportions were calculated from the sample proportions by use of the appropriate upper-tail probability. All statistical analyses were performed using MedCalc® for Windows (MedCalc Software).

Results

The perfusion defect sizes at SPECT, expressed as the percentage of left ventricle, ranged from 0% to 63% (median, 21%), and LVEF in the acute phase ranged from 17% to 58% (median, 40%). CK-MB peak concentrations ranged from 17 to 1323 μg/L, with a median value of 275 μg/L. We found a significant positive correlation between plasma cTnT concentrations (median, 2.27 μg/L; range, 0.04–7.55 μg/L) and both peak CK-MB ($r = 0.76$; $P < 0.001$; Fig. 1) and perfusion defect size at SPECT ($r = 0.62$; $P < 0.001$; Fig. 2). We also found a close correlation between peak CK-MB concentrations and scintigraphic defect sizes ($r = 0.66$; $P < 0.001$).

When we assessed the relationships between cTnT and the indices of LV function obtained by SPECT performed at the time of CCU discharge, cTnT was inversely correlated with LVEF ($r = -0.56$; $P < 0.001$; Fig. 3A) and positively correlated with LV end-systolic volume ($r = 0.58$; $P < 0.001$) and LV end-diastolic volume ($r = 0.55$; $P < 0.001$). cTnT >2.27 μg/L predicted a LVEF of <40% at CCU discharge with a sensitivity of 82.8% (95% CI, 64.2–94.1%), a specificity of 80.0% (95% CI, 63.1–91.5%), and a likelihood ratio for a positive test of 4.1 (95% CI, 3.7–4.7). ROC curve analysis produced an area under the curve (± SE) of 0.82 ± 0.05 (95% CI, 0.71–0.91; Fig. 4).

When we assessed the relationship between cTnT measured at CCU discharge and SPECT indices obtained 3 months after AMI, the overall accuracy of cTnT in predicting LV function increased significantly. cTnT remained significantly inversely correlated with LVEF ($r = -0.70$; $P < 0.001$; Fig. 3B), and the area under the ROC curve was 0.91 (95% CI, 0.80–0.97), indicating the high diagnostic accuracy of cTnT (Fig. 4). cTnT >2.98 μg/L predicted a LVEF of <40% at 3 months after AMI with a sensitivity of 86.7% (95% CI, 59.5–98.0%), a specificity of 81.4% (95% CI, 66.6–91.6%), and a likelihood ratio for a positive test of 4.7 (95% CI, 4.0–5.4). The results of a reanalysis to examine the effect of revascularization by excluding the 10 patients who did not receive reperfusion therapy were not different from the results for all patients.

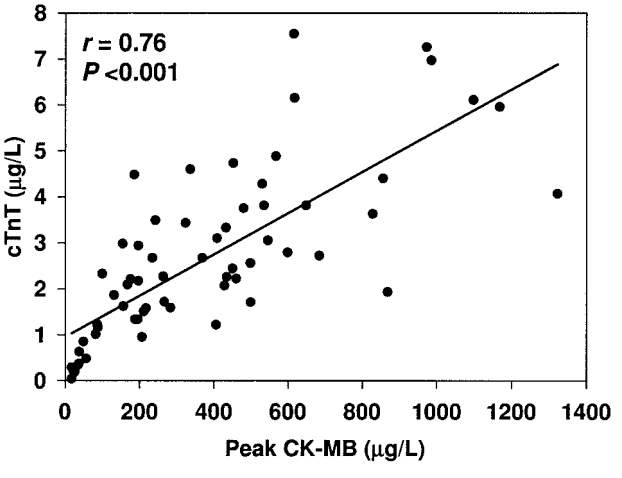

Fig. 1. Correlation between cTnT at CCU discharge and peak CK-MB concentrations in patients with AMI.

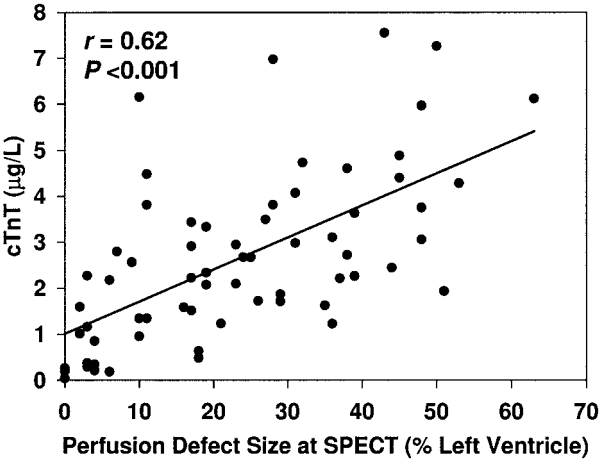

Fig. 2. Correlation between cTnT and the scintigraphic estimate of infarct size, both measured at CCU discharge, in patients with AMI.

Discussion

Estimation of the extent of myocardial injury from the release of CK-MB into plasma is a common clinical practice *(11)*. However, cardiac troponins have recently been proposed as a new biochemical standard for diagnosis of AMI; consequently, hospitals may want to consider the replacement of CK-MB with cardiac troponins *(20)*. One of the major concerns about replacing CK-MB with cardiac troponins is the lack of robust evidence on the ability of troponins to estimate the AMI size in clinical practice *(26)*. Few studies have evaluated troponins as markers for assessment of infarct size in comparison with traditional serologic markers and other techniques *(17, 18, 27)*. Consequently, we decided to evaluate the ability of cTnT to predict infarct size and LV function in patients with AMI admitted to CCU of our general hospital to support the definitive implementation of cTnT and the replacement of CK-MB in our laboratory cardiac panel. In particular, we evaluated the possibility of using a more advantageous single cTnT measurement, performed at approximately the time corresponding to the plateau phase of cTnT release in the blood, rather than previously used marker kinetics and peaks.

In our study, cTnT closely correlated with SPECT estimates of infarct size and peak serum concentrations of CK-MB. Our results corroborate those of earlier clinical studies in which cTnT release, not a single-point measure-

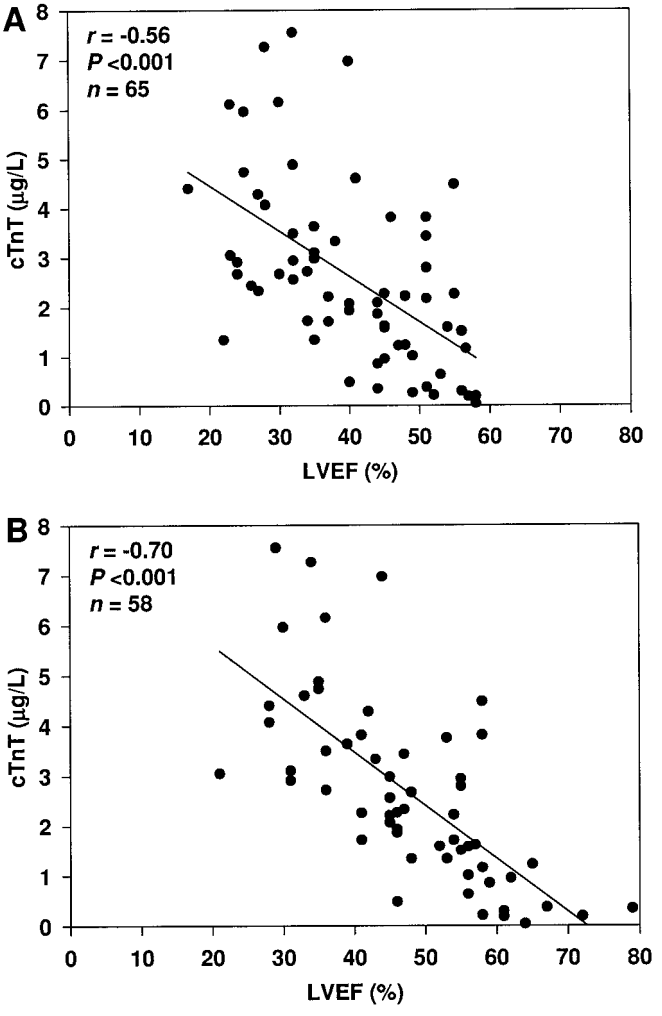

Fig. 3. Correlation between cTnT at CCU discharge and LVEF assessed at CCU discharge (*A*) and 3 months after hospitalization (*B*) in AMI patients.

ment, was used to assess infarct size *(17, 18)*. Furthermore, our findings in living patients are consistent with those obtained experimentally in dogs by Remppis et al. *(15)*, who found a good correlation ($r = 0.69$; $P = 0.003$; n = 16) between cTnT concentrations 96 h after the onset of ischemia and the pathoanatomic infarct size as quantified by the 2,3,5-triphenyltetrazolium chloride method. In clinical practice, estimation of AMI size based on cTnT determination on a single plasma sample at CCU discharge would facilitate the choice of appropriate care, leading to more efficient and economic use of healthcare facilities. This approach appears to be more useful than analyzing cumulative cTnT release, as proposed previously, because of the requirement of repetitive sampling and a possible incomplete recovery of cTnT *(28)*.

LVEF is a very powerful prognostic indicator after AMI *(23)*. A strong inverse relationship exists between LV function and patient outcome, with rapidly increasing mortality rates at LVEFs <40% *(24)*. Rao et al. *(29)* first showed a good correlation between cTnT concentrations measured 12–48 h after admission and LVEF ($r = -0.72$; $P < 0.001$; n = 50). In

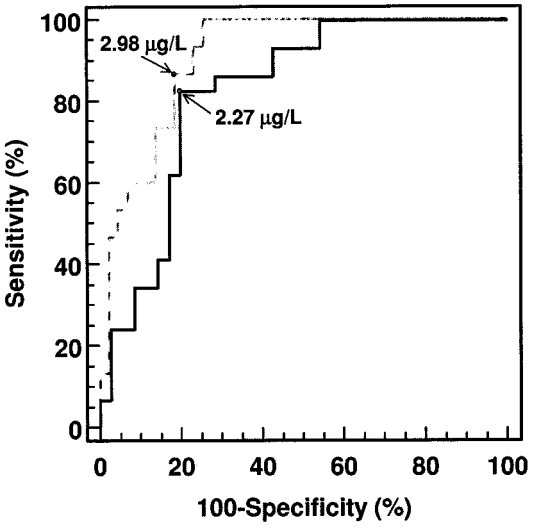

Fig. 4. ROC curve analysis for cTnT measured at CCU discharge as a diagnostic test of LVEF <40% in the acute phase of myocardial infarction (*black line*) and 3 months later (*gray line*).
Best decision thresholds are shown.

the study, a cTnT >2.8 μg/L predicted a LVEF <40% with a sensitivity of 100% and specificity of 93% (area under the ROC curve, 0.98) *(29)*. More recently, Kanna et al. *(30)* confirmed that serum cTnT on day 3 or 4 after AMI was significantly negatively correlated with LVEF assessed 1 month later ($r = -0.48$; $P < 0.001$; n = 86). No studies to date, however, have used gated SPECT imaging to assess LV function in comparison with cTnT. Our study is the first to demonstrate a significant inverse relationship between LVEF derived from gated SPECT and plasma cTnT on CCU discharge of post-AMI patients. We repeated the LVEF estimate ~3 months after the onset of AMI because this index can be affected during the acute phase by a combination of myocardial necrosis, ischemia, periinfarction edema, and stunned myocardium *(31)*. LV function and volumes may therefore not always represent true values in the acute phase after AMI, even if gated SPECT images are acquired at rest, as was done in our study, to guard against errors *(22)*. The increase in the overall accuracy of cTnT in predicting LV function, when estimated 3 months after AMI, reinforces our results. Although our patients with AMI were chosen without selection bias, the majority of them showed transmural necrosis, and all of them were admitted early. This increased the homogeneity of the population. Our patient group also experienced very few complications. This would further affect the results because complications associated with potential reinfarction or congestive heart failure would clearly confound the analysis. Therefore, our data apply only to a subset of patients who have transmural AMI, present early, and have no further complications. The proposed timing of sampling is appropriate only for this subset of patients. Those with small amounts of myocardial necrosis, those who receive later intervention, and those with complications are not classifiable based on our sampling protocol and corresponding results.

In conclusion, the present study shows a clear relationship between cTnT on CCU discharge and SPECT indices of infarct size and LV function in patients with a first AMI. If confirmed in larger and more heterogeneous patient populations, a single measurement of plasma cTnT on the day of CCU discharge after AMI, regardless of the kinetics of marker appearance in the blood, could be used as a convenient, cost-effective, noninvasive estimate of infarct size and for the assessment of LV function in a routine clinical setting. This may reveal a reliability similar to that for peak CK-MB concentrations (which, however, require repetitive sampling) or nuclear imaging (not available for all patients in the acute phase of AMI and too expensive to be used routinely).

References

1. Sobel BE, Bresnahan GF, Shell WE, Yoder RD. Estimation of infarct size in man and its relation to prognosis. Circulation 1972;46:640–8.
2. Vassanelli C, Menegatti G, Nidasio GP, Franceschini L, Cocco C, Rizzotti P. Comparison of different pharmacological interventions on enzymatic parameters during acute myocardial infarction. Clin Biochem 1987;20:441–7.
3. Christenson RH, Vollmer RT, Ohman EM, Peck S, Thompson TD, Duh SH, et al. Relation of temporal creatine kinase-MB release and outcome after thrombolytic therapy for acute myocardial infarction. Am J Cardiol 2000;85:543–7.
4. Hørder M, Hyltoft Petersen P, Thygesen K, Lyager Nielsen B. Plasma enzymes in myocardial infarction. An appraisal of quantitative, clinical and pathophysiological information. Scand J Clin Lab Invest 1981;41:41–7.
5. Holman BL, Goldhaber SZ, Kirsch CM, Polak JF, Friedman BJ, English RJ, et al. Measurement of infarct size using single photon emission computed tomography and technetium 99m pyrophosphate: a description of the method and comparison with patients prognosis. Am J Cardiol 1982;50:503–11.
6. Kaul S. Assessing the myocardium after attempted reperfusion. Should we bother? [Editorial]. Circulation 1998;98:625–7.
7. Grande P, Fischer Hansen B, Christiansen C, Naestoft J. Estimation of acute myocardial infarct size in man by serum CK-MB measurements. Circulation 1982;65:756–64.
8. Hackel DB, Reimer KA, Ideker RE, Mikat EM, Hartwell TD, Parker CB, et al. Comparison of enzymatic and anatomic estimates of myocardial infarct size in man. Circulation 1984;70:824–35.
9. Grande P, Christiansen C, Alstrup K. Comparison of ASAT, CK, CK-MB, and LD for the estimation of acute myocardial infarct size in man. Clin Chim Acta 1983;128:329–35.
10. tk;2Burlina A, Rizzotti P, Plebani M, Cocco C, Vassanelli C, Menegatti G. CPK and CPK-MB in the early diagnosis of acute myocardial infarction and prediction of infarcted area. Clin Biochem 1984;17:356–61.
11. Al-Saady NM, Camm AJ. New soluble markers for assessment of infarct size. In: Kaski JC, Holt DW, eds. Myocardial damage. Early detection by novel biochemical markers. Dordrecht, The Netherlands: Kluwer Academic Publishers, 1998:137–48.
12. Katus HA, Remppis A, Scheffold T, Diederich KW, Kuebler W. Intracellular compartmentation of cardiac troponin T and its release kinetics in patients with reperfused and nonreperfused myocardial infarction. Am J Cardiol 1991;67:1360–7.
13. Voss EM, Sharkey SW, Gernert AE, Murakami MM, Johnston RB, Hsieh CC, et al. Human and canine cardiac troponin T and creatine kinase-MB distribution in normal and diseases myocardium. Arch Pathol Lab Med 1995;119:799–806.
14. Ricchiuti V, Sharkey SW, Murakami MM, Voss EM, Apple FS. Cardiac troponin I and T alterations in dog hearts with myocardial infarction. Correlation with infarct size. Am J Clin Pathol 1998; 110:241–7.
15. Remppis A, Ehlermann P, Giannitsis E, Greten T, Most P, Müller-Bardorff M, et al. Cardiac troponin T levels at 96 hours reflect myocardial infarct size: a pathoanatomical study. Cardiology 2000;93:249–53.
16. Hammerer-Lercher A, Metzler B, Mair J, Pachinger O, Xu Q. Cardiac troponin T closely correlates with infarct size in a murine model of acute myocardial infarction [Abstract]. Eur Heart J 2001; 22(Suppl):674.
17. Wagner I, Mair J, Fridrich L, Artner-Dworzak E, Lechleitner P, Morass B, et al. Cardiac troponin T release in acute myocardial infarction is associated with scintigraphic estimates of myocardial scar. Coron Artery Dis 1993;4:537–44.
18. Tanaka H, Abe S, Yamashita T, Arima S, Saigo M, Nakao S, et al. Serum levels of cardiac troponin I and troponin T in estimating myocardial infarct size soon after reperfusion. Coron Artery Dis 1997;8:433–9.
19. Hallermayer K, Klenner D, Vogel R. Use of recombinant human cardiac troponin T for standardization of third generation troponin T methods. Scand J Clin Lab Invest 1999;59(Suppl 230):128–31.
20. Myocardial infarction redefined—a consensus document of The Joint European Society of Cardiology/American College of Cardiology Committee for the redefinition of myocardial infarction. J Am Coll Cardiol 2000;36:959–69.
21. Panteghini M, Gerhardt W, Apple FS, Dati F, Ravkilde J, Wu AH. Quality specifications for cardiac troponin assays. International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) Scientific Division—Committee on Standardization of Markers of Cardiac Damage. Clin Chem Lab Med 2001;39:174–8.
22. Bavelaar-Croon CDL, Pauwels EKJ, van der Wall EE. Gated single-photon emission computed tomographic myocardial imaging: a new tool in clinical cardiology. Am Heart J 2001;141:383–90.
23. The Multicenter Postinfarction Research Group. Risk stratification and survival after myocardial infarction. N Engl J Med 1983;309: 331–6.
24. Kroll D, Farah W, McKendall GR, Reinert SE, Johnson LL. Prognostic value of stress-gated Tc-99m sestamibi SPECT after acute myocardial infarction. Am J Cardiol 2001;87:381–6.
25. Swets JA. Measuring the accuracy of diagnostic systems. Science 1988;240:1285–93.
26. Wu AHB, Apple FS, Gibler WB, Jesse RL, Warshaw MM, Valdes R Jr. National Academy of Clinical Biochemistry standards of laboratory practice: recommendations for the use of cardiac markers in coronary artery diseases. Clin Chem 1999;45:1104–21.
27. Mair J, Wagner I, Morass B, Fridrich L, Lechleitner P, Dienstl F, et al. Cardiac troponin I release correlates with myocardial infarction size. Eur J Clin Chem Clin Biochem 1995;33;869–72.
28. Kragten JA, Hermens WT, van Dieijen-Visser MP. Cardiac troponin T release into plasma after acute myocardial infarction: only fractional recovery compared with enzymes. Ann Clin Biochem 1996;33:314–23.
29. Rao ACR, Collinson PO, Canepa-Anson R, Joseph SP. Troponin T measurement after myocardial infarction can identify left ventricular ejection of less than 40%. Heart 1998;80:223–5.
30. Kanna M, Nonogi H, Sumida H, Miyazaki S, Daikoku S, Morii I, et al. Usefulness of serum troponin T levels on day three or four in predicting survival after myocardial infarction. Am J Cardiol 2001;87: 294–7.
31. Braunwald E, Kloner RA. The stunned myocardium: prolonged, postischemic ventricular dysfunction. Circulation 1982;66:1146–9.