Accuracy of Myocardial Biomarkers in the Diagnosis of Myocardial Infarction After Revascularization as Assessed by Cardiac Resonance: The Medicine, Angioplasty, Surgery Study V (MASS-V) Trial

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Background. The lack of a correlation between myocardial necrosis biomarkers and electrocardiographic abnormalities after revascularization procedures has resulted in a change in the myocardial infarction (MI) definition.

Methods. Patients with stable multivessel disease who underwent percutaneous or surgical revascularization were included. Electrocardiograms and concentrations of high-sensitive cardiac troponin I (cTnI) and creatine kinase (CK)-MB were assessed before and after procedures. Cardiac magnetic resonance and late gadolinium enhancement were performed before and after procedures. MI was defined as more than five times the 99th percentile upper reference limit for cTnI and 10 times for CK-MB in percutaneous coronary intervention (PCI) and coronary artery bypass grafting (CABG), respectively. Sensitivity of cTnI was 100%, 100%, and 100% and of CK-MB was 96.4%, 96.4%, and 96.4%, respectively. The best cutoff of cTnI for on-pump CABG, off-pump CABG, and PCI patients was 21.7%, 28.3%, and 52.4% and for CK-MB was 72.5%, 81.2%, and 90.5%, respectively. The specificity of cTnI was 3.6%, 9.4%, and 42.1% and of CK-MB was 73.2%, 86.8%, and 96.4%, respectively. The best cutoff of cTnI for on-pump CABG, off-pump CABG, and PCI was 6.5 ng/mL, 4.5 ng/mL, and 4.5 ng/mL (162.5, 112.5, and 112.5 times the 99th percentile upper reference limit) and of CK-MB was 37.5 ng/mL, 22.5 ng/mL, and 11.5 ng/mL (8.5, 5.1, and 2.6 times the 99th percentile upper reference limit), respectively.

Conclusions. Compared with cardiac magnetic resonance, CK-MB was more accurate than cTnI for diagnosing MI. These data suggest a higher troponin cutoff for the diagnosis of procedure-related MI.

The Joint European Society of Cardiology/American College of Cardiology/Federation of European Societies for Cardiology/American Heart Association/World Heart Federation Task Force proposed a redefinition for acute myocardial infarction (AMI) [1]. This redefinition was motivated by the emergence of more sensitive necrosis biomarkers, such as sensitive cardiac troponin I (cTnI), which is currently referred to as a contemporary “sensitive” assay that is capable of detecting small changes in myocardial viability and necrosis after interventional procedures. Thus, cTnI is considered the preferred biomarker for detection of myonecrosis. However, the writing committee noted that these definitions were arbitrarily chosen, of uncertain

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clinical relevance, and not grounded on substantial scientific evidence linking their occurrence to subsequent adverse outcomes [1]. The isolated use of biomarkers to detect small areas of injury failed to clearly identify AMI based on the gold standard [2]. In this scenario, the use of cardiac magnetic resonance (CMR) to confirm or dismiss the event appears to be appropriate. Considering these conflicting data, this study aimed to determine the accuracy of cardiac biomarkers in the diagnosis of MI after revascularization procedures using the new late gadolinium enhancement (LGE) technique in CMR and to rate these results based on the third definition of AMI by the Joint European Society of Cardiology/American College of Cardiology Foundation/American Heart Association/World Heart Federation Task Force [1].

Patients and Methods
Details of the Medicine, Angioplasty, Surgery Study V (MASS-V) study design, protocol, patient selection, and inclusion criteria have been previously reported [3]. Briefly, patients with angiographically documented proximal multivessel coronary stenosis of more than 70% by visual assessment and documented ischemia were included. Ischemia was documented by stress testing or the stable angina assessment of the Canadian Cardiovascular Society (class II or III). All patients were candidates for elective percutaneous coronary intervention (PCI) or coronary artery bypass grafting (CABG). Patients were excluded if they experienced any or all of previous mechanical interventions, recent thromboembolic phenomena, systemic inflammatory disease, or kidney failure.

Trial Outcomes
The primary outcome was the occurrence of MI with the release of biomarkers, cTnI and creatine kinase (CK)-MB, together with LGE detected by CMR.

CMR Protocol
CMR was chosen as the gold standard because it allows a highly accurate and reproducible assessment of myocardial injuries within the same examination. Recent studies indicate that CMR for detection of MI is accurate [4] and provides results similar or superior to radionuclide imaging [5].

All patients in this trial were forwarded to CMR 2 days before the intervention and 6 days after each invasive procedure during the hospitalization period. The patients were studied in a 1.5-T Achieva MR scanner (Philips Healthcare, Andover, MA). Steady-state free-precession cine images were acquired in 2 long-axis (2 and 4 chambers) views and 8 to 10 short-axis views of the left ventricle.

Contrast-enhanced images were acquired in long- and short-axis planes identical to the cine images. Typical voxel size was 1.6 x 2.1 x 8 mm, with a reconstruction matrix of 528 and a reconstructed voxel size of 0.6 mm. MI was defined as the identification of hyperenhancement in the myocardium on CMR. Infarcted regions exhibit this phenomenon, which might be due to an increased volume of distribution of the contrast agent because of rupture of myocyte membranes and slow contrast washout [6].

CMR Analysis
All areas of late gadolinium-diethylene triamine pentaacetic acid hyperenhancement were quantified by 2 experienced observers who interpreted the LGE while blinded to the interventional technique and biochemical data. When measurements differed, a third observer performed a review, and a consensus was obtained. Hyperenhanced pixels were defined as those with image intensities exceeding 2 standard deviations greater than the mean of image intensities in a remote myocardial region in the same image. Preintervention and post-intervention scans were read side by side in both surgical techniques, with and without extracorporeal circulation.

Measurement of cTnI and CK-MB
Blood samples were collected for measurement of cTnI and CK-MB mass immediately before PCI and 6, 12, 24, 36, and 48 hours after. For patients undergoing on-pump (ONCAB) or off-pump CABG (OPCAB), these cardiac markers were measured immediately before and 6, 12, 24, 36, 48, and 72 hours after the operation.

The treating surgeon and clinical team were blinded to the CK-MB or cTnI data. All samples were centrifuged at 3,000 rpm for 20 minutes and analyzed within 2 hours after collection. Analyses of cTnI and CK-MB were performed using an ADVIA Centaur immunoassay analyzer (Siemens Health Care Diagnostics, Tarrytown, NY). According to the manufacturer, the lower limit of detection of cTnI using the high-sensitivity Ultra kit is 0.006 ng/mL, and the 99th percentile upper reference limit (URL) is 0.04 ng/mL. The assay precision represented by the percentage coefficient of variation is 10% at 0.03 ng/mL. The detection limit of the CK-MB mass kit is 0.18 ng/mL. Cutoff values at the 99th percentile are 3.8 ng/mL for women and 4.4 ng/mL for men. The coefficient of variations for CK-MB mass, as specified by the manufacturer, are 3.91% at 3.55 ng/mL and 3.67% at 80.16 ng/mL. These measurements are in accordance with the recommendations of the Study Group on Biomarkers in Cardiology of the European Society of Cardiology Working Group on Acute Cardiac Care [7].

Definition of CABG-PCI–Related AMI
According to the Third Universal Definition [1], AMI type 4a for PCI and type 5 for CABG are defined as an elevation of more than five times the 99th percentile URL during the first 48 hours after PCI (in patients with normal baseline cTnI concentration) plus any of the following: (1) evidence of prolonged ischemia as demonstrated by prolonged chest pain, (2) ischemic ST-segment changes or new pathologic Q waves, (3) angiographic evidence of a flowing-limiting complication, or (4) imaging evidence of a new loss of viable myocardium or new regional wall motion abnormality; and 10 times the 99th percentile URL for CABG during the first 48 hours.
after CABG (in patients with normal baseline cTnI concentrations) plus any of the following: (1) new pathologic Q waves or new left bundle branch block (LBBB), (2) angiographically documented new graft or new native coronary occlusion, or (3) imaging evidence of new loss of viable myocardium or new regional wall motion abnormality.

**Electrocardiograms**

Twelve-lead ECGs were obtained from each patient immediately before and 6, 12, 24, and 36 hours after PCI and CABG. For the identification of new Q waves, we used the Minnesota code, which is used extensively in epidemiology studies and large-scale clinical trials [8].

**Ethics Committee Approval**

All patients provided written informed consent and were assigned to a treatment group. The Ethics Committee of the Heart Institute of the University of São Paulo Medical School, São Paulo, SP, Brazil, approved the trial. All procedures were performed in accordance with the Declaration of Helsinki.

**Statistical Analysis**

Values are expressed as the mean ± standard deviation or median (interquartile range), as appropriate. The paired-sample t test and the unpaired-sample t test were used to compare means within the study group or between subgroups. The $\chi^2$ statistics with the Fisher exact test was used for comparison of discrete variables. Continuous variables without normal distribution were compared with the Mann-Whitney U test, and correlation between such variables was made with the Spearman rank test.

Binary logistic regression was performed to determine which clinical and angiographic parameters predicted the likelihood of myocardial hyperenhancement. Multivariate analysis was also performed and adjusted for age, sex, diabetes mellitus, hypertension, treatment allocation, previous MI, smoking status, number of diseased vessels, and a positive treadmill test (variables associated with poor outcomes) based on the presence of new hyperenhancement after PCI or CABG. Assessment of the association between the ECG and biomarkers was performed using the agreement correlation coefficient $\kappa$. Areas under the receiver operating characteristic curves were calculated and compared using MedCalc 13.0 software (MedCalc Software, Mariakerke, Belgium). The DeLong method was used to determine optimum cutoff thresholds. Values of $p < 0.05$ were considered statistically significant.

**Results**

Between May 2012 and March 2014, 326 prospective, nonrandomized patients were eligible for isolated CABG or PCI in a single center, and 219 were included in this trial. The main reasons for the exclusion of 107 patients are presented in Figure 1. Of the included patients, 148 were assigned to the CABG group, and 71 were assigned to the PCI group. In the CABG group, 73 patients were assigned to undergo OPCAB, and 75 were assigned to undergo ONCAB; Fig 1). The baseline characteristics of the patients were similar in the three groups (Table 1). Objective evidence of myocardial ischemia was observed in 173 patients (79%). Triple-vessel disease was present in 137 patients (62%), with 92% involvement of the anterior descending artery.
CABG was performed in 136 patients, in whom 544 distal anastomoses were also performed, averaging 3.4 anastomoses per patient. PCI was performed in 211 lesions in 66 patients, averaging 3.1 stents per patient. No significant differences occurred in major in-hospital complications between the groups. No overall in-hospital deaths, fatal MIs, or in-hospital reoperations occurred in any study patient. Multivariate analysis of the variables mentioned above identified no association with the emergence of new hyperenhancement after PCI or CABG. The main data related to biomarkers, ECG, and LGE-CMR are presented in Table 2.

ECG Results

Of the 69 patients in the ONCAB group, 67 (97.1%) had no Q wave ECGs compared with the baseline ECG. The remaining 2 patients had a new Q wave together with the release of necrosis biomarkers and delayed enhancement on CMR, reaching the concordance index of 0.972; 95% CI, 0.983 to 0.992. Enzymatic elevation in the presence of LGE was found in all patients with Q wave ECGs.

CMR Results

Of the 69 patients in the ONCAB group, CMR in 56 (81%) was unchanged compared with that at baseline. The remaining 13 patients (19%) had delayed enhancement on CMR. Furthermore, 53 of the 67 OPCAB patients (79%) had an unchanged CMR compared with that at baseline. The remaining 14 patients (21%) had delayed enhancement on CMR. In addition, 57 of the 66 PCI group patients (86.3%) had an unchanged CMR compared with that at baseline, and the remaining 9 (13.7%) had evidence of delayed enhancement on CMR.

Serum Biomarkers of Myocardial Injury

A total of 2,706 cTnI and CK-MB samples were obtained from the CABG and PCI patients, yielding a mean of 13.3 samples per biomarker per patient. In the CABG group, cTnI elevation of cardiac biomarker values exceeding 10 times the 99th percentile URL were observed in 129 patients (94.8%); only 7 (5.2%) had values of these biomarkers within the normal reference range. However, CK-MB mass with elevation of cardiac biomarker values exceeding 10 times the 99th percentile URL was observed in 40 patients (29.4%); 96 patients (70.6%) had values for these biomarkers within the normal reference range. The
PCI group data show that cTnI elevation of cardiac biomarker values exceeding five times the 99th percentile URL occurred in 42 patients (64%), and only 24 patients (36%) had values within the normal reference range. Regarding CK-MB, 5 patients (7.6%) had elevated cardiac biomarker values exceeding five times the 99th percentile URL, and 61 (92.4%) had values within the normal reference range. Table 2 reports the accuracy and predictive values for CABG and PCI patients. Figure 2 illustrates the best cutoff values in the area under the curve.

**Comment**

Different correlations between troponin and CK-MB with myocardial infarct size have been demonstrated in a histopathologic study [9]. The highest values of troponin exhibited poor correlations with the extent of infarction. By contrast, the highest serum levels of CK-MB exhibited poor correlations with the extent of infarction. Despite this, the highest values of troponin exceeded three times the 99th percentile URL as 100%, 93%, and 60%, respectively, demonstrating that cTnI is overly sensitive and identifies many patients without objective evidence of CMR-detected myocardial injury. Although small cTnI elevations may represent myonecrosis not detectable by CMR, the prognostic significance of such small elevations has not been demonstrated. The authors also proposed that changing the cTnI threshold for MI diagnosis to 40 times the 99th percentile URL would greatly enhance specificity (93%) without reducing sensitivity (100%). This 13-fold bioequivalence ratio of cTnI to CK-MB is even greater than the sevenfold ratio reported from the Evaluation of Drug-Eluting Stents and Ischemic Events (EVENT) registry, in which a cTnI threshold exceeding 20 times the upper limit of normal was associated with similar mortality rates as a CK-MB increase exceeding three times [11]. These data confirm that very different thresholds for CK-MB and cTnI after revascularization must be considered to represent similar levels of myonecrosis [12]. Compared with our study, CK-MB was strictly within the limits established by the Third Universal Definition, whereas cTnI was beyond this boundary.

The application of the Third Universal Definition of Myocardial Infarction to interventional procedures in our study revealed that CK-MB was superior to cTnI for myocardial necrosis not detectable by CMR, the prognostic significance of such small elevations has not been demonstrated.

**Table 2. Biomarkers, Images, and Procedure-Related Variables**

<table>
<thead>
<tr>
<th>Variables</th>
<th>ONCAB (n = 69)</th>
<th>OPCAB (n = 67)</th>
<th>PCI (n = 66)</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>cTnI Abnormal</td>
<td>67</td>
<td>62</td>
<td>42</td>
<td></td>
</tr>
<tr>
<td>Normal</td>
<td>2</td>
<td>5</td>
<td>24</td>
<td>0.368</td>
</tr>
<tr>
<td>CK-MB Abnormal</td>
<td>24</td>
<td>16</td>
<td>5</td>
<td></td>
</tr>
<tr>
<td>Normal</td>
<td>45</td>
<td>51</td>
<td>61</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>CMR No enhancement</td>
<td>56</td>
<td>53</td>
<td>57</td>
<td></td>
</tr>
<tr>
<td>Enhancement</td>
<td>13</td>
<td>14</td>
<td>9</td>
<td>0.588</td>
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<tr>
<td>Electrocardiogram No Q wave</td>
<td>67</td>
<td>62</td>
<td>65</td>
<td></td>
</tr>
<tr>
<td>Q wave</td>
<td>2</td>
<td>5</td>
<td>1</td>
<td>0.684</td>
</tr>
</tbody>
</table>

* Abnormal (values >10 times the 99th percentile upper reference limit for coronary artery bypass grafting and >5 times 99th percentile upper reference limit for PCI); normal (values <10 times the 99th percentile upper reference limit for CABGs and <5 times 99th percentile URL for PCI).

**Table 3. Comparison of the Diagnostic Accuracy of Cardiac Biomarkers Between Late Gadolinium Enhancement-Cardiographic Magnetic Resonance, Electrocardiogram, and Coronary Intervention**

<table>
<thead>
<tr>
<th>Variables</th>
<th>ECG</th>
<th>CK-MB</th>
<th>cTnI</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(%)</td>
<td>(%)</td>
<td>(%)</td>
</tr>
<tr>
<td>Sensitivity</td>
<td>NPCAB</td>
<td>OPCAB</td>
<td>PCI</td>
</tr>
<tr>
<td>ONCAB</td>
<td>15.3</td>
<td>61.5</td>
<td>100</td>
</tr>
<tr>
<td>OPCAB</td>
<td>35.7</td>
<td>57.1</td>
<td>100</td>
</tr>
<tr>
<td>PCI</td>
<td>11.1</td>
<td>44.4</td>
<td>100</td>
</tr>
<tr>
<td>Specificity</td>
<td>ONCAB</td>
<td>OPCAB</td>
<td>PCI</td>
</tr>
<tr>
<td>ONCAB</td>
<td>96.4</td>
<td>73.2</td>
<td>3.6</td>
</tr>
<tr>
<td>OPCAB</td>
<td>92.4</td>
<td>86.8</td>
<td>9.4</td>
</tr>
<tr>
<td>PCI</td>
<td>91.2</td>
<td>96.4</td>
<td>42.1</td>
</tr>
<tr>
<td>PPV</td>
<td>ONCAB</td>
<td>OPCAB</td>
<td>PCI</td>
</tr>
<tr>
<td>ONCAB</td>
<td>100</td>
<td>37.5</td>
<td>19.4</td>
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<tr>
<td>OPCAB</td>
<td>100</td>
<td>56.2</td>
<td>22.6</td>
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<tr>
<td>PCI</td>
<td>100</td>
<td>80.0</td>
<td>23.1</td>
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<td>NPV</td>
<td>ONCAB</td>
<td>OPCAB</td>
<td>PCI</td>
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<tr>
<td>ONCAB</td>
<td>80.6</td>
<td>91.1</td>
<td>100</td>
</tr>
<tr>
<td>OPCAB</td>
<td>79.3</td>
<td>90.2</td>
<td>100</td>
</tr>
<tr>
<td>PCI</td>
<td>87.7</td>
<td>91.4</td>
<td>100</td>
</tr>
<tr>
<td>Accuracy</td>
<td>ONCAB</td>
<td>OPCAB</td>
<td>PCI</td>
</tr>
<tr>
<td>ONCAB</td>
<td>78.3</td>
<td>72.5</td>
<td>21.7</td>
</tr>
<tr>
<td>OPCAB</td>
<td>76.1</td>
<td>82.1</td>
<td>28.3</td>
</tr>
<tr>
<td>PCI</td>
<td>77.2</td>
<td>90.5</td>
<td>52.4</td>
</tr>
</tbody>
</table>

* Values in parentheses indicate the number of patients treated according to the gold standard/total number of patients.

CK-MB = creatine kinase-MB; CMR = cardiac magnetic resonance; cTnI = cardiac troponin I; ONCAB = on-pump coronary artery bypass; OPCAB = off-pump coronary artery bypass; PCI = percutaneous coronary intervention; ECG = electrocardiogram; PPV = positive predictive value.
identifying myocardial necrosis using CMR as the gold standard. When comparing the accuracy between the two biomarkers using CMR, we observed that CK-MB was more accurate than cTnI in both CABG and PCI patients. All patients with hyperenhancement on CMR had elevated biomarkers and myocardial perfusion defects related to the target vessel; however, hyperenhancement was not observed in patients without elevated biomarkers.

Our study further observed a strong specificity between the appearance of new Q waves on ECG and delayed enhancement on CMR, even though ECG has decreased sensitivity for myocardial necrosis [13]. In this same scenario, data from a randomized clinical trial revealed no correlation between ECG and CMR findings [14]. Only 6% of patients undergoing surgical treatment demonstrated new Q waves, and 39% of these patients had new late enhancement on CMR.

The release of myocardial necrosis markers in the absence of MI may occur frequently during surgical revascularization. This phenomenon is probably due to the nonphysiologic effects of cardiopulmonary bypass (CPB), undocumented venous thromboembolism, coagulation disturbances after CPB, and ventricular aneurysmectomy. In addition, during PCI, the release of enzymes likely due to occlusion of the collateral circulation and the no-reflow phenomenon or distal microembolization, or both, are frequently observed.

Cellular changes during cardiac or systemic pathophysiologic disturbances may promote the release of these enzymes. Thus, the temporary injury of myocytes and the subsequent release of enzymes and their recovery must not necessarily be construed as indicative of necrosis or an AMI. After myocardial injury, a portion of the troponin content is hypothesized to be liberated directly from the cytosolic pool, but most is retained longer because it is associated with the contractile apparatus. Thus, the serum elevation is slow and irregular in contrast with CK-MB, which is exclusively derived from the cytosol [15].

The release of such biomarkers is strongly related to the extent of trauma, regardless of the occurrence of AMI. Indeed, measurement of the increase in trauma level indicates that there is a linear increase and progressive release of biomarkers, especially CK-MB. We observed that the release of biomarkers that accounted for the best cutoff for cTnI for PCI, OPCAB, and ONCAB was 112.5, 112.5, and 162.5 times the 99th percentile URL, respectively, and was 2.6, 5.1, and 8.5 times the 99th percentile URL for CK-MB, respectively.

Data recently published in a small trial of 40 surgical patients show a release of biomarkers exceeding 10 times the 99th percentile URL for both cTnI and CK-MB; however, only 8 patients had delayed enhancement on CMR [16]. The lack of correlation between necrosis biomarkers and CMR was identified in a recent study [17] of patients who underwent CABG and PCI. Using the Third Universal Definition of AMI in the PCI patient group, the authors found that 75% of patients exhibited cTnI release, and only 15.8% had delayed enhancement on CMR ($p = 0.001$). When they evaluated the CABG group, 100% exhibited cTnI release, and only 28.1% had delayed enhancement on CMR ($p = 0.0001$). The following limitations of this study should be considered in the interpretation of our findings. Our results cannot be generalized to other cTnI assays given that different cTnI assays are not biologically equivalent because manufacturers use different reference populations to determine the 99th percentile URL. Moreover, this study was conducted in a single center, and its results can be regarded as limited to this center. However, this study provides a more homogeneous sample. Finally, the sample studied can be considered too small to provide definitive results. Although our statistical data are robust, multicenter
studies with specific goals are needed to establish the best cutoff for biomarkers.

**Clinical Implications**

Because cardiac biomarker testing is recommended by The Joint European Society of Cardiology, American College of Cardiology Foundation, American Heart Association, and World Heart Federation Task Force, it is essential that further research be directed toward understanding the clinical relevance of proteins. Simply measuring cTnI or CK-MB concentrations for AMI is only the tip of the iceberg. The identification and understanding of disease-induced contractile protein modifications and their clinical significance will have an ultimate effect on future assay design.

**Conclusions**

Assuming that necrosis biomarkers have limited diagnostic accuracy in myocardial injury, the challenges faced for the establishment of definitive values for the diagnosis of myocardial damage include new cutoff values for cTnI. Furthermore, the diagnosis of this condition cannot be exclusively based on cardiac biomarkers or ECG. It is reasonable to include CMR in the set of tools for the accurate diagnosis of procedure-related myocardial injury.

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**References**


