Troponin I Measurement after Myocardial Infarction and its Correlation with Left Ventricular Ejection Fraction: A Prospective Study

Deepak Somani***, RS Gahlot*, Manoj Lakhotia**, Chaina Ram Choudhary***, Sanjeev Sangavi**

Abstract

Objective: To determine the relationship of serum troponin I after first acute myocardial infarction with left ventricular ejection fraction as assessed by echocardiography.

Methods: A total of 50 patients of acute myocardial infarction were included in the study. Troponin I concentration was measured by ELISA method and echocardiographic ejection fraction was calculated by modified Simpson's rule. Echocardiographic ejection fraction was compared with serum troponin I concentration. Patients with previous myocardial infarction were excluded.

Result: There was strong negative correlation between troponin I concentration and left ventricular ejection fraction, i.e., with an increasing troponin level, there was a fall in ejection fraction. The Pearson's correlation coefficient was –0.69, which was statistically significant (p < 0.0001). It was also found that cardiac troponin I (cTnI) concentration > 6.6 ng/ml predicted LVEF < 50% with a sensitivity of 100% and specificity of 92.4%.

Conclusion: Serum troponin I concentration has a strong negative correlation with left ventricular ejection fraction after first acute myocardial infarction, and hence can be used to assess the LVEF in patients with first myocardial infarction. A level of 6.6 ng/ml provided a good indication for LVEF below 50% and thus can identify patients with higher risk.

Key words: Troponin I, Left ventricular ejection fraction, Myocardial infarction.

Introduction

After acute myocardial infarction (AMI), a patient's prognosis is closely related to the extent of irreversibly damaged myocardium. In routine clinical practice, infarct size is estimated non-invasively by electrocardiography, imaging techniques (such as myocardial radionuclide imaging and echocardiography), and serological tests.

There is an increasing awareness of the limitations of standard biochemical markers of cardiac damage in patients with AMI. A desire to improve sensitivity, specificity, and prognostic value has led to the search for markers uniquely expressed by the myocardium. Troponin is a globular protein of muscle that binds to tropomyosin and has a marked affinity for calcium ions, and is thus a central regulatory protein of muscle contraction. The troponin, a protein-complex, consists of three subunits with different structure and functions (T, I, C). Troponin I is a 23.5 kDa component of Troponin complex that inhibits the interaction of myosin cross-bridges with the actin–tropomyosin complex, and thus regulates the striated muscle contraction. Three isoforms of Troponin I are present: slow twitch and fast twitch skeletal muscle isoforms, and cardiac muscle isoform.

The cardiac Troponin I (cTnI) has been found to have excellent sensitivity and specificity and is superior to creatine kinase–MB (CK-MB) as indicator of myocardial necrosis. cTnI is uniquely located in the myocardium and its release closely relates to infarct size; therefore, inversely correlates with left ventricular ejection fraction (LVEF). We performed this study to find out the level of cTnI after AMI, and its correlation with LVEF.

Research design and methods

This prospective study included 50 patients of AMI admitted in the CCU. Myocardial infarction was diagnosed if at least two of the following criteria were present: cardiac chest pain, ST segment elevation of at least 2 mm in chest leads or 1 mm in limb leads, and raised creatine kinase MB activity. Patients with significant renal impairment, rheumatoid arthritis, and a history of heart failure (due to any cause), and previous myocardial infarction, were excluded. No control subjects were taken because cTnI...
is not detected in the peripheral circulation under normal circumstances.

Serum troponin I concentration was measured between 12 - 48 hours after the onset of chest pain by enzyme-linked Immuno sorbent assay using polyvinyl microtitre plates (micro-ELISA).

The assay system utilises the four monoclonal antibodies directed against the cTnI. Three mouse monoclonal anti-troponin I antibodies are used for solid phase immobilisation (on the microlitre wells). The fourth antibody is in the antibody enzyme (horse radish peroxide) conjugate solution. The test sample is allowed to react simultaneously with the four antibodies resulting in the troponin I molecule being sandwiched between the solid phase and enzyme-linked solution. After 90 minutes of incubation at room temperature, the wells are washed with water to remove unbound labelled antibodies. A solution of TMB reagent is added and incubated for 20 minutes, resulting in the development of a blue colour. The colour development is stopped with the addition of 3 N HCl, changing the colour to yellow. The concentration of Troponin I is directly proportional to the colour intensity of the test sample. Absorbance is measured spectrophotometrically at 450 nm.

At present there is no WHO standardisation for Troponin I due to the use of different Troponin I antibodies in the test. In our study, the test kit was by BIO-CHECK Inc. made in Burlingame CA. It was calibrated with Abbot's Ax Sym Troponin I test. The diagnostic cut-off for the AMI patient was determined to be 2 ng/ml.

Echocardiograms were obtained using a GE-Ving Med System V echocardiographic machine (GE-Ving Med sound AB, Horten, Norway) with a 3.5 mHZ multiphase array probe in subjects lying in the left lateral decubitus position and supine position. The echocardiographic techniques and calculations of different cardiac dimensions were performed according to the recommendations of the American Society of Echocardiography.

The ejection fraction was obtained using a modified biplane Simpson's method from apical two chamber and four chamber view (LV2D). Measurements were made from three consecutive beats, and the average of three beats was used for analysis. LVEF less than 50% was taken as systolic dysfunction.

**Statistical analysis**

The relation between LVEF and cTnI concentration was studied using Pearson’s correlation coefficient, and by systemic analysis of sensitivity and specificity. Patients were initially categorised into two data sets, those with EF < 50% and those with EF > 50%. This ejection fraction value was based on previous clinical trials as having prognostic significance. Student ‘t’ test was used to compare means of variable in two groups.

**Results**

Table I shows the clinical feature and biochemical characteristic of the study group. Mean age of study group was 55 + 9 years with 34 (68%) being male. Most common risk factor was dyslipidaemia (54%) followed by hypertension (32%).

<table>
<thead>
<tr>
<th>Variables</th>
<th>Values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (yrs)</td>
<td>55 + 9</td>
</tr>
<tr>
<td>Sex (M/F)</td>
<td>34/16</td>
</tr>
<tr>
<td>Dyslipidaemia (%)</td>
<td>54</td>
</tr>
<tr>
<td>Hypertension (%)</td>
<td>32</td>
</tr>
<tr>
<td>Diabetes Mellitus (%)</td>
<td>20</td>
</tr>
<tr>
<td>Smoking (%)</td>
<td>36</td>
</tr>
<tr>
<td>Alcohol (%)</td>
<td>28</td>
</tr>
<tr>
<td>Cholesterol (mg/dl)</td>
<td>197 + 30.6</td>
</tr>
<tr>
<td>Serum Triglyceride (mg/dl)</td>
<td>174.7 + 72.9</td>
</tr>
<tr>
<td>Serum LDL (mg/dl)</td>
<td>117.4 + 25.4</td>
</tr>
<tr>
<td>Serum HDL (mg/dl)</td>
<td>39.14 + 6.7</td>
</tr>
</tbody>
</table>

Table II shows the relation between cTnI and LVEF. There was a strong negative correlation between cTnI level and LVEF. The pearson’s correlation coefficient between cTnI and LVEF was r = -0.69. The cTnI value was high among patients with LVEF < 50%. The difference was statistically significant (p < 0.0001).
Table II: cTnI levels (mean ± SD) in relation to ejection fraction.

<table>
<thead>
<tr>
<th>Ejection fraction</th>
<th>n</th>
<th>Trop. I ng/ml (mean ± SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 50%</td>
<td>24</td>
<td>11.49 ± 6.34</td>
</tr>
<tr>
<td>≥ 50%</td>
<td>26</td>
<td>5.07 ± 1.48</td>
</tr>
</tbody>
</table>

(p < 0.0001)

Table III shows the sensitivity and specificity of cTnI to predict LVEF. It was found that cTnI concentration > 6.6 ng/ml predicted LVEF of < 50% with a sensitivity of 100% and specificity of 92.4%.

Table III: cTnI level and ejection fraction.

<table>
<thead>
<tr>
<th>Ejection fraction</th>
<th>Troponin I</th>
<th>&lt; 50%</th>
<th>&gt; 50%</th>
</tr>
</thead>
<tbody>
<tr>
<td>(n)</td>
<td>(a)</td>
<td>(b)</td>
<td></td>
</tr>
<tr>
<td>≥ 6.6</td>
<td>24</td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>&lt; 6.6</td>
<td>0</td>
<td>23</td>
<td></td>
</tr>
</tbody>
</table>

a = true positive  c = false negative
b = false positive d = true negative

Discussion

cTnI is accepted as a highly reliable biochemical marker for detecting myocardial damage, and its use in the diagnosis of acute myocardial infarction is increasing. Data shows that cTnI is related to the amount of myocardial damage, but there are very few studies to substantiate the claim. cTnI release closely relates to infarct size and therefore inversely correlates with left ventricular ejection fraction, as there is inverse relation between infarct size and left ventricular ejection fraction.

This study shows a strong negative correlation between cTnI concentration measured 12 - 48 hours post-myocardial infarction and echocardiographic left ventricular ejection fraction (r = -0.69, p < .0001). It was also found that cTnI concentration > 6.6 ng/ml is a sensitive (100%) and specific (92.4%) indicator of left ventricular ejection fraction of < 50% after a first myocardial infarction. These results were similar to the results obtained by Sharkey et al.

One limitation of the study was that 2 D calculation of LVEF was done, which is always smaller than that determined by angiography, and thus chances of technical errors are there. However, it can still be said that levels of CK-MB mass (upper reference limit 5 ng/ml) and cTnI (upper reference limit 0.8 ng/ml using Dade Stratus II assay) in serial serum specimens obtained over 36 hrs after the onset of chest pain from patients of AMI. cTnI was increased significantly (p < 0.05) over CK-MB after 9-12 hours. Sixteen of twenty patients assessed by echocardiography had an abnormal left ventricular ejection fraction (LVEF) mean 37.6 (SD-15.2%), ranging from 15.4 to 67.6%. LVEF was significantly inversely correlated to peak CK-MB (r = 0.5, p < 0.03) as well as to cTnI (r = 0.46, p < 0.04). Similarly, Hara et al. studied the relationship between cTnI, various biochemical markers, and left ventricular ejection fraction (LVEF) after successful direct percutaneous transluminal coronary angioplasty (PTCA) in 36 patients with AMI. Biochemical markers were measured on admission, immediately after, and from 6 hours to 9 days after PTCA. The time to peak values were CK-MB 9.7 hours, cTnI 9.8 hours, cTnI 18.6 hours, and myosin light chain 68.9 hours. LVEF inversely correlated with peak values of CK-MB (r = -0.519, P < 0.01) cTnI (r = -0.500, P < 0.01) cTnI (r = -0.44, p < 0.05) and myosin light chain (r = -0.441, p < 0.05). The value of cTnI at each sampling point was significantly inversely related to chronic phase LVEF.

So, based on these findings, cTnI shows excellent promise as a marker of infarct size, and for the assessment of LVEF; and may potentially replace the CK-MB as the cardiac specific marker for AMI detection.

cTnI has practical advantages over other markers in the assessment of left ventricular ejection fraction. After acute infarction, cTnI has a peak value at 12 hours from the onset of pain. The plateau phase of cTnI, however, lasts up to 48 hours, and represents an integrated estimate of myocyte necrosis. The peak value will therefore be missed in samples taken 12 - 48 hours after admission, but there is a large time window. This makes repeated sampling unnecessary, and represents a cost and time-effective method of diagnosis and quantification. This is in contrast to creatine kinase-MB or myoglobin, for which multiple measurements are required to identify the peak value and whose values are affected by thrombolysis.

One limitation of the study was that 2 D calculation of LVEF was done, which is always smaller than that determined by angiography, and thus chances of technical errors are there. However, it can still be said that levels of...
cTnI provide a reasonable approach to select patients with left ventricular dysfunction who may require further interventions.

This marker offers a simple, inexpensive, quick non-invasive method of identifying such patients. Estimation of troponin I can also be used to identify those patients who may benefit from other treatments, for example, ACE inhibitors.

References