

Cardiac Myosin-Binding Protein C as a new biomarker in the diagnosis of myocardial infarction

Abstract

Myocardial infarction is an emergency situation, and its early diagnosis is vital for patients and for the treatment which will be followed. Till now the diagnosis of myocardial infarction is based on the increased cardiac troponins I and T in the patient's serum, these biomarkers are the gold standard for the diagnosis of myocardial infarction but there are some limitations due to their slow release from the damaged myocytes. For this reason, new biomarkers are needed to be used for a faster diagnosis. New biomarkers should increase rapidly after acute myocardial infarction and should have cardiac selectivity. Myosin-binding protein C is a newly described candidate biomarker of cardiac injury and in small studies, it appears that its serum concentration decreases and increases faster than that of the troponin T and I.

Cardiac troponins are released and cleared slowly after myocardial injury, complicating the diagnosis of an early and recurrent, acute myocardial infarction. Cardiac myosin-binding protein C is a similar cardiac protein that may have different release/clearance kinetics.

Keywords: Myocardial infarction, Acute myocardial infarction, Cardiac myosin binding protein C, troponin T, Cardiac myosin binding protein C, and myocardial infarction

1. Introduction

Chest pain is a non-specific symptom and it is responsible for 6-10% of patients' attendance to the emergency department. Early diagnosis of acute myocardial infarction (AMI) is vital as early treatment can reduce the high mortality .[1-3]

The diagnosis of patients is a difficult task as only 32% of patients with AMI have diagnostic changes in the electrocardiogram (ECG), such ST interval elevation or ST interval depression, which has a high prognostic value for AMI. However, only 10% of these patients have a final diagnosis of AMI. Consequently, the identification of patients with AMI depends almost entirely on the measurement of the systemic circulation of cardiac troponin (cTn) I or cTn (T) released by the damaged myocardium. However, these biomarkers have the disadvantage that they are released slowly and require repeated blood tests. Many patients have "positive troponin", but not necessarily be "AMI positive" .[3-6] The obstruction of a coronary artery due to the formation of plaques on its inner walls leads to a reduced supply of oxygen to the myocardium resulting in necrosis of myocardial cells .[7]

The cause of myocardial infarction (MI) could be due to acute rupture of the atherosclerotic plaque and thrombus formation, type 1 MI [T1MI] (STEMI or N-STEMI), or could be due to the changes in myocardial oxygen supply or / oxygen demand in the absence of acute atherothrombosis, type 2 MI [T2MI]. Type 2 MI is not common enough, and in most cases, it is caused by conditions that increase myocardial oxygen demand (tachyarrhythmia, arterial hypertension, excessive fatigue) or conditions that reduce oxygen supply (coronary artery spasm) or due to reduced oxygen throughout the body (e.g., severe anemia, severe chronic respiratory

disease) .[8] The ECG shows lesions such as ST-elevation (STEMI) although sometimes it may occur as non-STEMI (NSTEMI) .[9] The AMI is accompanied by laboratory evidence of tissue damage. The biomarkers used for verification are cardiac troponin (cTn), and creatine kinase-MB (CK-MB) .[9]

The European guidelines recommend the measuring of hs-cTnI and hs-cTnT for the rapid exclusion of AMI. The concentrations which are used are well below the 99th percentile set by the population (for exclusion) and above the 99th percentile to significantly improve the exclusion sensitivity .[10,11]

1.1 Troponin (cTn)

Troponin was introduced as an indicator of cardiac necrosis in cardiology in the late 1987s by Cummins et al. Troponin I or T (cTnI and cTnT) are commonly used to confirm or rule out the diagnosis of AMI, or in suspected acute coronary syndromes without ST-elevation (NSTEMI-ACS) .[12-14]

Troponins (cTns) are a group of proteins that help regulate stimulation-contraction of the cardiac and skeletal muscles. Responding to three isoforms, Troponin C (TnC) which binds calcium, troponin I (TnI) inhibits the ATPase activity of the actomyosin complex, and it is an important regulator of myosin-actin interactions in the contraction and relaxation of the cardiomyocytes. Troponin T (TnT) binds to tropomyosin .[13,15-17]

The types of cardiac troponin I (cTnI) and T (cTnT) are different from those of troponin cTnI and cTnT which are found in the skeletal muscles. Troponin C is found in slow skeletal muscle, and it is not considered a biomarker of cardiac necrosis. When there is damage to myocardial cells, cardiac troponin I and T are released into

the circulation. The greater the extent of the lesion, the greater the concentration of troponin I and T .[15,16,18] The biological elimination,the half-life of cTns is relatively short (cTnT: 120 minutes, cTnI: 90 minutes) and the time needed to see the increased levels of cTns in the plasma depends on the etiology of the heart failure. When a patient develops AMI, cTns show a specific kinetic profile, troponin levels in the blood rise within 3-12 hours and may remain elevated for 7 and 14 days .[19]

Over the past decade, there have been significant improvements in the sensitivity and accuracy of cTn analysis, to reduce the time required to detect an increase in its levels. Although cardiac troponins are used to detect acute ischemic damage, they may be elevated in several other heart conditions, such as myocarditis, atrial tachycardia , [13,15,20] and various non-cardiac conditions such as sepsis and endotoxemia, burns, as well as respiratory diseases such as pulmonary embolism. Elevated cTn levels may occur in healthy athletes after strenuous exercise. Patients with chronic kidney disease also show a consistently low increase in the levels of troponin .[21-25]

From the above, it is clear that cTns have potential deficiencies and new necrosis biomarkers could prove valuable. New biomarkers should increase faster after acute myocardial damage and must have cardiac selectivity .[14,26]

2. Cardiac myosin-binding protein C (cMyC)

Myosin-binding protein C is recently described as a new candidate biomarker for cardiac injury, and it was shown in small group studies that its serum concentration increases and decreases faster than that of troponin T and I .[5,6]

Cardiac myosin-C binding protein (cMyBP-C) is a cardio-specific regulatory protein that plays a role in the regulating of the cardiac contraction, and diastolic relaxation of cardiomyocytes .[27,28] As with cTnT and cTnI, cMyC expression is restricted to the heart but in higher concentrations. In addition, cMyCs increase more rapidly in systemic circulation after AMI than hs-cTnT, and this may be due to their higher concentration in the myocardium.[29]

2.1 Structure of Cardiac myosin-binding protein C (cMyC)

The sarcomeric protein, the cardiac protein that binds myosin C (C-protein, MYBPC3, cMyBP-C, or cMyC), is thick and it is one of the most abundant cardiac protein. The cMyC is found in three isoforms in adult human muscle: slow skeletal, fast skeletal, and cardiac muscles encoded by different genes (encoded by MYBPC1 and MYBPC2 genes on chromosomes 12q23.3 and 19q33.3, respectively), and a cardiac isoform (cMyBP-C, MYBPC3 gene on chromosome 11p11.2) .[29-32]

Cardiac isoform differs from skeletal isoforms by having an extra immunoglobulin-like region at the N-terminus (C0), phosphorylation sites between regions C1 and C2, and an introduction of 28 amino acids into region C5 and expressed exclusively in the heart .[29,31]

The cMyBP-C consists of eleven subunits from C0 to C10, eight Ig-like (IgC2 like) and three fibronectin (FN3) (Fn (III) -like) type domains with a proline-alanine-rich region between C0 and C1, and a link between the region C1 and C2 (i.e., M-domain).(33-35) Each cMyBP-C molecule binds via the C-terminus to the dense myosin filament, while its N-terminal domains are free to come into contact with either the myosin tip or the thin actin filament. In addition to myosin binding, cMyBP-C interacts with actin through the N-terminal regions (C0 - C2). These

partner-binding interactions are thought to functionally affect cardiac contractility .[34,36,37]

Several observations in mice have shown that the loss of cMyBP-C has inhibitory effects on the myocytes, thus slowing down the time course of ventricular relaxation .[38] In addition to its inhibitory effects, cMyBP-C may also activate actomyosin interactions .[39] This is due to the ability of the cMyBP-C N'-terminal regions to bind the actin and can shift the tropomyosin to its open state in the thin strand .[28,40]

The contraction of the cardiac muscle begins with the activation of thin fibers that contain actin but are formed by structural changes in the dense filaments that contain myosin. Calcium-binding to the troponin causes the removal of tropomyosin from the actin-binding regions of myosin, which allows the myosin transverse bridge from adjacent thick filaments to bind to actin .[41] Both inhibitory and activating interactions of cMyBP-C are thought to be controlled by the phosphorylation status of cMyBP-C, which appears to be essential for normal myocardial function and appears to protect against ischemic injury .[26]

The phosphorylation of cMyBP-C in the Ser-273, Ser-282, and Ser-302 and Ser-307 regions appears to regulate myocardial contractile function and provide resistance to proteolysis, thus maintaining cardiac function after AMI. All four phosphorylation sites are located in the myosin between the C1 and C2 regions .[42,43] Loss of phosphorylation has often been associated with heart failure, atrial fibrillation, or cardiomyopathy, further emphasizing their functional importance .[44,45]

2.1.2 Troponin T and cMyC concentration in patients with acute myocardial infarction (AMI)

There are few studies in the literature which are comparing the release and serum concentration of cMyBP-C concerning the release and concentration of troponin (cTn). The results of these studies are encouraging, and it appears that the serum cMyBP-C concentration increases and decreases more rapidly than that of troponin T and I.

An experimental study by James O. Baker compared the maximum release time of cMyC with that of cTnT in serum samples from 20 patients with STEMI, and a cTnT concentration 12 hours after introduction of 2000 ng / L. The results of the study showed that the serum concentration of cMyC in patients with myocardial injury increased and decreased more rapidly relative to cTnT levels. However, the study had some limitations as it did not include patients with NSTEMI-ACS, but patients with documented myocardial damage .[46]

In another study by Gonvindan et al. cMyBP-C levels were determined concerning current diagnostic biomarkers (cTnI, MYO, CK-MB, GP-BB, H-FABP (CAIII, carbonic anhydrase III; CK-MB, creatine kinase-MB; cMyBP-C, cardiac myosin binding protein-C; GP-BB, glycogen phosphorylase BB; H-FABP, heart-type fatty acid-binding protein I) by immunosorbent assay in 65 patients with AMI before and after Percutaneous Transcatheter Coronary Intervention (PTCA) compared with levels of 54 healthy controls. The results showed that compared with healthy subjects (22.3 ± 2.4 ng / mL), plasma cMyBP-C levels were significantly increased in patients with AMI (105.1 ± 8.8 ng / mL) as well as a significant reduction in the 12 hours after PTCA (41.2 ± 9.3 ng / mL) compared with patients with MI. Plasma cMyBP-C

measurement showed 66.2% sensitivity and 100% specificity. These findings showed that cMyBP-C is a sensitive cardiac biomarker with potential utility for the accurate diagnosis of AMI .[47] These findings are consistent with another study also conducted by Kuster et al. where it was found that cMyC circulates faster than cTnT. This study investigated the release of myosin cardiac binding protein-C (cMyBP-C) compared to existing cTnI, cTnT, and myosin light chain-3 (MYL3) biomarkers in both animal and human models. In an animal model in 6 pigs, plasma cMyBP-C levels were measured 30 minutes to 14 days after coronary artery occlusion. Coronary plasma levels of cMyBP-C peaked at 6 hours after coronary ligation. The levels of cTnI, cTnT, and myosin light chain-3 levels all increased 6 hours after ligation.

In the same study in 12 patients with hypertrophic obstructive cardiomyopathy who underwent transcatheter ablation of septal hypertrophy, cMyBP-C increased significantly and peaked at 4 hours. Also, in the same study in 176 patients with NSTEMI, serum levels of cMyBP-C were significantly higher than those of the control group of 153 subjects. This study showed that cMyBP-C in all 3 groups was released into the blood rapidly after myocardial injury in the first 3 hours, peaked at 6 hours, and returned to baseline values at 12 hours. In contrast, cTnI, and cTnT levels were significantly lower within the first 3 hours and increased at later timepoints .[48]

In another study by Kaie et al. kinetic release of cMyC was described using quantitative immunoassay in 20 patients with STEMI who underwent therapeutic removal of septal hypertrophy (TASH) for hypertrophic cardiomyopathy or in coronary artery bypass graft (CABG). In both MI models (STEMI, TASH), the results showed an early increase in cMyC compared to a high sensitivity cTnT assay and

faster cMyC disappearance. In the same study, cMyC was analyzed in 1954 selected patients with AMI symptoms in emergency departments in a prospective, diagnostic multicenter study, and the performance of cMyC was compared with that of hs-cTnT and hs-cTnI. AMI was the final diagnosis in 340 patients (17%), and a much greater potential range of cMyC was observed in patients with AMI compared with patients without AMI and compared with both hs-cTn assays.

Also, in 174 patients with suspected AMI, who presented with chest pain very early after the onset of their symptoms, lasting less than 3 hours, the concentration of cMyC / hs-cTnI in blood samples was taken at three different time points. The results showed a positive linear correlation between the two biomarkers. However, the mean ratios in patients with AMI were much higher at presentation than at later times (median 2.72 at 0 hours, 1.83 at 3 hours, 0.63 at 6-12 hours), suggesting a more dynamic increase in cMyC in the early stages of myocardial infarction than hs-cTnI. The results may be since cMyC is found in higher concentrations in myocardial cells or a different mechanism of protein release from damaged myocardial cells.[4]

In a prospective diagnostic study by Kaier et al. cMyC and high-sensitivity cardiac troponin were compared. Blood samples were taken by ambulance-based paramedics in patients with chest discomfort, acute dyspnea in the absence of known lung disease. Sampling was performed within the first 70 minutes after the onset of symptoms. The study included 776 patients of whom 22% were diagnosed with AMI. The results showed that the cMyC concentration in patients with AMI was significantly higher than in patients with other diagnoses. The diagnosis for AMI was better with cMyC than with high-sensitivity cardiac troponin T. The study showed that the new biomarker excels in the early diagnosis of patients with AMI or other

diseases, based on the analysis that used a prognostic approach such as the application and comparison of regression accounting models. It has also shown that cMyC may have distinct advantages as a biomarker for AMI and that a cMyC concentration <10 ng / L may be sufficient after 2 hours of symptoms to reliably rule out AMI. This advantage of cMyC is obvious despite the use of hs-cTnT in the final diagnosis of AMI.[49]

The APACE study included patients >18 years old who came to the emergency departments with symptoms of acute chest pain that started within the first 12 hours. Patients with end-stage renal disease and hemodialysis were excluded. The study also excluded patients with MI with an elevation of the ST segment. A blood sample was then taken (at time points of 1 hour, 2 hours, 3 hours, and 6 hours). The study included a total number of 1954 unselected patients, of whom 1469 patients did not have significant ECG abnormalities on arrival at the hospital. The mean time from onset of chest pain was 5 hours. A total number of 340 patients were diagnosed with AMI, unstable angina in 10%, symptoms of cardiac origin other than coronary heart disease in 14%, non-cardiac symptoms in 54%, and symptoms of unknown origin in 5%. The cMyC levels were significantly higher in patients with AMI compared to patients with other diagnoses such as unstable angina. Similarly, the concentrations of cTnT, hs-cTnI, and s-cTnI in the blood were significantly higher in AMI compared to other final diagnoses. Overall, blood concentrations of cMyC relative to the limit of detection were higher than those of hs-cTn in all diagnostic categories.

In this multicenter diagnostic study, the diagnostic performance of cMyC was compared with that of cTnI and cTnT, using the major hs assays recommended in the current guidelines in patients with symptoms indicative of AMI. The results showed

that the cMyC concentration was higher than hs-cTnT in patients with <3 hours of the onset of chest pain .[50] Therefore cMyC can be a valuable biomarker in patients who come to the emergency department early after the onset of their symptoms.[50]

3. Conclusion

Cardiac necrosis biomarkers have become vital in confirming or excluding AMI as well as in suspected acute coronary syndromes with non-ST elevation (NSTE-ACS). Cardiac troponins are the gold markers in the diagnosis of MI. However, they have the disadvantage that they increase slowly in the serum, resulting in many times the delayed diagnosis. Myosin-binding protein (cMyC) is a newly described biomarker of cardiac necrosis. Myosin binding protein C (cMyC) is released more rapidly into the circulation than cardiac troponins cTnT and cTnl, following AMI, due to its higher concentration in myocardial cells. Its diagnostic accuracy for AMI is higher than that of cardiac troponin T and I, and especially in patients with onset of chest pain <3 hours. This results in the early diagnosis of patients with AMI.

In conclusion, the results of the studies suggest that serum cMyC levels increase and decrease significantly relative to cTnT levels in patients with AMI, even in the first hours after the onset of symptoms, playing an important role in their medical treatment of the patients. Therefore, cMyC as a new biomarker of myocardial necrosis should be further investigated.

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