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REVIEW ARTICLE

Cardiac Biomarkers in Acute Coronary Syndrome

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ABSTRACT

Acute coronary syndrome (ACS) is one of the leading causes of admission to the emergency departments (EDs) worldwide. The diagnosis of ACS involves the evaluation of clinical signs and symptoms, electrocardiographic assessment, and measurement of cardiac circulating biomarkers. In the last 60 years, the use of laboratory markers has changed considerably. Early biomarker assessment has entailed testing for total enzyme activity of aspartate aminotransferase (AST), lactate dehydrogenase (LDH), and creatine kinase (CK) but was highly nonspecific. Soon thereafter, the development of immunoassays, as well as technical advances in automation, allowed the measurements of the CK MB isoenzyme (CK-MB) in mass rather than in activity and myoglobin. Currently, cardiac troponins (CTn) have the highest sensitivity and specificity for myocardial necrosis and represent the biochemical gold standard for diagnosing acute myocardial infarction (AMI). This review provides a chronology of the major events that marked the evolution of cardiac biomarker testing and the development of the relative assays from the first introduction of AST in the 1950s to the last high-sensitivity troponin immunoassays in the 2010s.

Keywords: Acute coronary syndrome, Acute myocardial infarction, Cardiac biomarkers, Cardiac troponin.

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INTRODUCTION

Coronary artery disease (CAD) is the leading cause of death among adults and one of the most common reasons for ED visits. Although the standard 12-lead electrocardiogram (ECG) is the single best test to identify patients with acute ST segment elevation myocardial infarction (STEMI) upon ED presentation, it still has relatively low sensitivity (only 35–50%) for detection of AMI.[1] Cardiac biomarkers are measurable and quantifiable biological parameters which are detected in the blood and serve as indices of assessments of AMI. In a subject having chest pain along with ECG changes, the elevation of cardiac biomarkers helps to diagnose non-ST segment elevation MI (NSTEMI) and STEMI.[2] The Joint European Society of Cardiology/American College of Cardiology[3] has proposed criteria for the diagnosis of AMI as shown in Table 1. It is estimated that 34% of all ACS events are repeat events, in line with recent data from the Global Registry of Acute Coronary Events (GRACE).[4] Coronary artery disease has assumed an epidemic proportion in India. Over 80% of deaths and 85% of disability from cardiovascular disease (CVD) occur in low- and middle-income countries. India is often considered to be the region with highest burden of CVD.[5] An interesting fact is that CAD affects Indians with greater frequency and at a younger age as compared with the developed countries, as well as many other developing countries.

WHY DO WE NEED BIOMARKERS FOR DIAGNOSIS?

Acute coronary syndrome is the result of numerous pathophysiological events like

- Plaque rupture with acute thrombosis,
- Progressive mechanical obstruction,
- Inflammation,
- Secondary unstable angina, and
- Dynamic obstruction (coronary vasoconstriction).

The cardiac biomarkers now help the physicians to understand the degree of inflammation, myocyte necrosis, vascular damage, and hemodynamic stress contributing to ACS. These levels of biomarkers noninvasively demonstrate the pathogenic changes in the heart (Graph 1). As seen in Graph 1, the detection of cTn in the blood of patients with ACS is indicative of myocardial necrosis with the presence of intracoronary thrombus and distal embolization of platelet microaggregates.[6] Necrosis

Table 1: Criteria for diagnosis of AMI[3]

Clinically typical rise and gradual fall [troponin (at least value above the 99th percentile of the upper reference limit, URL)] or more rapid rise and fall (CK-MB) of biochemical markers with at least one of the following:
• Ischemic symptoms
• Development of pathologic Q waves on ECG
• Electrocardiographic changes indicative of ischemia (ST segment elevation or depression)
• Coronary artery intervention (e.g., coronary angioplasty)

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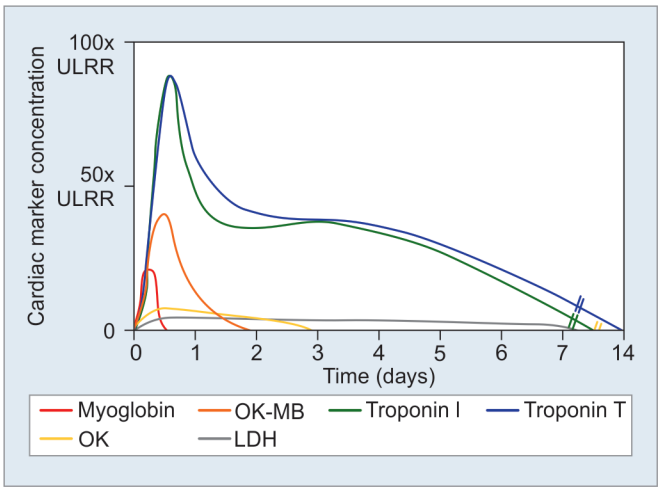

Graph 1: Cardiac markers—time *vs* levels post-MI[6]

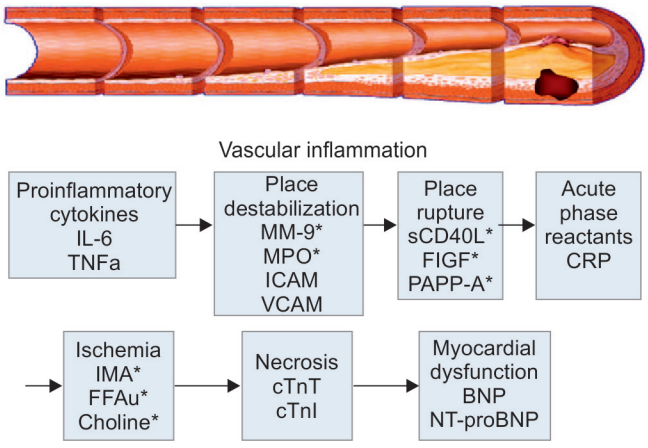

Fig. 1: Markers of inflammation, ischemia, and necrosis in heart[10]

compromises the integrity of the sarcolemmal membrane; intracellular macromolecules (serum and plasma cardiac markers) begin to diffuse into cardiac interstitium and ultimately into microvasculature and lymphatics. The rate of appearance of these macromolecules in the peripheral circulation depends on several factors, including intracellular location, molecular weight, local blood and lymphatic flow, and the rate of elimination from the blood.[7,8] It is important to clarify that these tests of myocardial injury do not provide any direct insight to the cause of the damage; it may be due to nonischemic insults.[9] These sensitive biomarkers (Fig. 1) guide the clinician in early management of myocardial ischemia to prevent necrosis with treatments, such as fibrinolysis, coronary artery bypass grafting, and percutaneous coronary interventions (PCIs) for improving outcomes.[10]

Criteria for IDEAL markers for MI

- *Specific*: It should be specific to myocardial muscle cells (no false positive)
- *Sensitive*: It should release rapidly on onset of attack (diagnose early cases)
  - It should be able to detect even minor damage
  - It should not miss positive cases (no false negative)
- *Prognostic*: Its level should relate with extent of damage
- *Persists longer*: It should stay longer in blood so that it can diagnose delayed admissions.
- *Reliable*: Procedure should depend on evidenced principle.
- *Simple, inexpensive*: It can be performed anywhere in low costs and does not need any highly qualified personnel.
- *Quick*: It should have low turnaround time. Results should be quicker, i.e., preferably within 30 minutes (maximum 1 hour) of hospital admission.

Types of Biochemical Markers for MI

- Cardiac enzymes (isoenzymes):
  - Total CK
  - CK-MB activity
  - LDH
  - AST
- Cardiac proteins:
  - Myoglobin
  - TnI and TnT
  - Pro-brain natriuretic peptide (pro-BNP)
- Nonspecific markers:
  - Leukocytosis (total leukocyte counts)
  - Raised erythrocyte sedimentation rate levels
  - Serum lipids
  - C-reactive protein (CRP)
  - Hemoglobin levels
- Newer markers

CARDIAC ENZYMES

1954: AST

- In 1954, serum glutamic oxaloacetic transaminase, now termed as AST, was identified as the very first biochemical marker for diagnosis of AMI.[11,12]
- AST increases in blood 3 to 4 hours after AMI, reaches the maximum value in blood in 15 to 28 hours, and returns to normal values within 5 days.[13] However, despite the high sensitivity for AMI, AST is a nonspecific biomarker of cardiac tissue, wherein its activity can also increase in several other conditions including hepatic congestion secondary to congestive heart failure, myocarditis, electrical cardioversion, pericarditis, tachyarrhythmias, pulmonary embolism, and shock.[14]

1955: LDH

- A cytoplasmic enzyme found in skeletal, muscle, liver, heart, kidney, and red blood cells. It is not a

tissue-specific enzyme. These are five izoenzymes, composed of four subunit peptides. Isoenzyme Type I is heart specific.[15,16] The ratio of LDH isoenzymes 1 and 2: the ratio is over 1.0 in AMI patients, whereas it remains below 1.0 in samples of patients with hemolysis.[17] Because of its prolonged half-life, LDH-1 is a clinically sensitive (90%) marker for infarction when measured after 24 hours.

- Hill and Levi[18] were the first to demonstrate the presence of LDH in human blood serum, and 1 year later Wróblewski and LaDue[19] and Wróblewski et al[20] observed an increase in LDH activity in serum of patients with AMI. Ulmer et al[21] confirmed this observation in a study population of 22 AMI patients.
- LDH and its isoenzyme LDH-1 increase in blood 5 to 10 hours after AMI, reach the maximum value in blood in 60 to 144 hours, and return to normal values in 12 days.[13]

1960: total creatinine kinase levels (sum of CK-MM, CK-MB, and CK-BB)

- It is a cytoplasmic and mitochondria enzyme of all body muscles. Thus, it is nonspecific to cardiac tissue (available in skeletal muscles also). It was in 1960 that the CK activity was shown to be a potential biomarker of cardiac muscle injury.[22]
- It gets elevated in 4 to 6 hours, peaks in 24 hours, and returns to normal values in approximately 72 hours[13]; the sensitivity of this biomarker is very high when blood is collected early after the onset of disease. Sorensen reported a sensitivity of 98% in the AMI diagnosis when blood was collected within 72 hours after the onset of disease.[23] Moreover, he also demonstrated that patients with high CK activity measurement in the third day had a worse prognosis.
- Years later, it was shown that total CK activity may be related to the extent of MI and prognosis.[24,25]
- Various types are
  - CK-1 (BB): Brain
  - CK-2 (MB): Myocardium
  - CK-3 (MM): Skeletal muscle, heart
- Limitations—nonspecific; it also gets elevated in any muscle disease, skeletal trauma, alcohol intoxication, seizures, vigorous exercise, thoracic outlet syndrome, kidney disease, and pulmonary embolism.

1972: CK-MB isoenzyme activity/mass

- In 1972, Roe et al[26] developed a zone electrophoresis method for the identification and quantification in serum or plasma of the CK-MB isoenzyme.
- The CK-MB isoenzyme, which is normally undetectable or very low in the blood, increases in both heart and skeletal diseases by showing highest concentration in cardiac muscle (~22% of the total CK content of myocardium compared with ~1 to 3% in the skeletal muscle).[27]
- Several studies confirmed that CK-MB subforms provide a reliable and specific diagnosis with high accuracy in the first hours of onset of cardiac symptoms.[28-30] Creatine kinase MB isoenzyme isoforms exist in only one form in cardiac muscle (CK-MB2) but exists in different isoforms in the plasma (CK-MB1).
- The mass method of CK-MB levels (that use monoclonal antibodies directed against CK-MB) has proved to be more accurate than traditionally radioimmunoassay or agarose gel electrophoresis methods, especially in patients presenting within 4 hours of injury. A study by Seo et al[31] compared CK-MB mass *vs* CK-MB activity and concluded that CK-MB mass is more sensitive when CK-MB concentration is in the low range.
- Puleo et al[28] reported a sensitivity and specificity of 95.7 and 93.9% respectively, with a high positive predictive value and a high negative predictive value within 6 hours of infarction. It is more specific than total CK. Creatine kinase MB isoenzyme (CK-2) has the most specificity for cardiac muscle (>85%), as normal skeletal muscle contains only 1% CK-MB. But is less specific than TnI. Creatine kinase MB isoenzyme appears in blood within 4 to 6 hours of onset of attack, peaks in 12 to 24 hours, and returns to normal within 2 to 3 days.
- One small study has shown a very high death rate (64%) during 4 years follow-up in patients who were admitted to the cardiac care unit with chest pain, positive CK-MB mass, and nondiagnostic ECG changes of STEMI. Several investigators have also studied the release of CK-MB mass following PCI.[32] They demonstrated that CK-MB mass is a sensitive indicator of myocardial injury following PCI. One study reported that 40% of patients showed evidence of myocardial damage following PCI using both CK-MB mass and cTnT.[33,34]
- Advantages: It is useful for early diagnosis of MI.
  - It is useful for diagnosis of reinfarction
- Disadvantages: It is not used for delayed admission (more than 2 days)

It is not 100% specific (elevated in skeletal muscle damage, strenuous exercise in long distance runners).

Pearson et al in 1990, proposed use of CK-MB relative index (CK-MB/total CK) to improve its specificity. Ratios greater than 2.5% are considered suggestive of myocardial damage. Also, a level of MB2 > 1 U/L, and a ratio of MB2/MB1 > 2.5 indicates myocardial injury at 6 hours.[35]

CARDIAC PROTEINS

1978: myoglobin

- Myoglobin is an iron- and oxygen-binding protein found in muscle tissue. It is only found in the blood stream when it is released following muscle injury.
- In 1978, myoglobin was detected for the first time.[36,37] It is a sensitive marker for muscle injury making it a potential marker for MI. It is not much specific for cardiac tissue (also found in skeletal muscles and renal tissue).
- Time sequence after infarction: It rises as fast as 2 hours, peaks at 6 to 8 hours, and returns to normal in 20 to 36 hours.
- Advantages: It has a high negative predictive value in early phase.

  It also has been used in assessing reperfusion after thromobolysis.

  In a study by Bhayana et al,[38] myoglobin was found to be superior to CK-MB mass and TnT for ruling out AMI within the period of 3 to 6 hours after symptom onset.
- Disadvantages: Not for delayed admission cases (after 1 day of onset of attack)

  Have false positives with skeletal muscle injury and renal failure.

1971: troponin isoforms

- In 1971, Greaser and Gergely demonstrated that the troponin complex actually consists of three components which were named TnC, TnI, and TnT in light of their specific properties: Ca^{2+} binding capacity (TnC), inhibition of ATPase activity (TnI), and tropomyosin binding respectively (TnT).[39]
- Cardiac-TnT (34 kDa) was first introduced in 1989 as a marker for AMI.[40]
- Complex of three protein subunits: (Fig. 2)
  1. TnC: Calcium-binding component
  2. TnI: Inhibitory component
  3. TnT: Tropomyozin-binding component
- cTn are different from skeletal muscle troponins. They are more specific for MI diagnosis (100% cardiac specific).

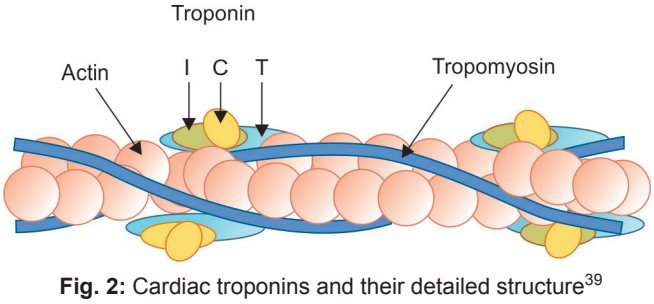

Fig. 2: Cardiac troponins and their detailed structure[39]

- cTn have a greater sensitivity for diagnosing minor damage of MI. It also increases in acute decompensated heart failure.
- Duration after MI
  - Appears in 2 to 6 hours
  - Peak in 12 to 16 hours
  - Stays elevated for 5 to 14 days
  - Early peaking in reperfusion
- Normal values

  TnT—0.1 to 0.2 ng/mL

  TnI—0.6 ng/mL
- Value rises to 20 to 50 times URL (99th percentile of reference control group). Patients with STEMI who undergo successful recanalization of the infarct-related artery have a rapid release of cTn, which can indicate reperfusion.
- Disappears from blood after about 1 week (stays longer). So, useful for diagnosis of delayed admission cases.
- Prognostic marker—there is a relation between its level in blood and extent of cardiac damage. Regardless of the etiology of troponin release, an elevated level is associated with worst prognosis.[41,42] An elevated troponin level indicates a higher-risk subgroup independent of ECG presentation and predischarge exercise testing.[43] There is an incremental risk of death or MI in patients with elevated troponins that can be seen in a quantitative fashion, even in patients with chronic renal insufficiency. Despite using the latest generation assays and higher cut-off values (higher than the 99th percentile) cTnT and cTnI were positive in 82 and 6% of dialysis patients respectively.[44] Even patients in whom CK-MB levels are within normal limits, troponin elevation signifies an increased risk of death when compared with those without elevation.[43]
- Serial measurements of cTn significantly improve the ability of this biomarker in detecting AMI.[45] Hamm et al[46] performed troponin testing upon presentation and at 4 hours with improvement in the sensitivity from 51 to 94% for cTnT and 66 to 100% for cTnI in patients without ST elevation. Higher diagnostic sensitivity and specificity require specimen collection at patient presentation, 6 to 9 hours later and at 12 to 24 hours if clinical suspicion is high and earlier results are negative.
- Elevated cTnT concentration has been reported in a significant number of patients with chronic renal failure. These levels do not seem to be affected by hemodialysis, with elevations persisting after treatment.[47] In chronic kidney disease (CKD) patients on hemodialysis without ACS, cTnI was reported to have better sensitivity and specificity for the diagnosis

of AMI compared with cTnT, but positive cTnT was associated with increased all-cause mortality during follow-up.[48,49]

- Elevated cTnT in patients with non-STEMI is associated with poor prognosis.[43] The increase correlates well with the severity of coronary artery lesions determined by angiography.[50] In one important study, the prognostic value of cTnT was assessed in 967 patients with unstable angina. It was found that the group that had elevated concentrations of cTnT had an increased risk of cardiac events, and the higher the cTnT, the more frequent the complications. Patients were followed up for 6 months for cardiac complications. The risk of further AMI and death was 4.3% in patients with cTnT < 0.06 µg/L and 16.1% for those with cTnT ≥ 0.18 µg/L.[51] In another study by Stubbs et al,[52] 62 non-STEMI patients were followed up for about 3 years after their admission with cTnT concentration ≥0.2 ng/mL. The incidence of complications in this group was very high: cardiac death (12 patients), coronary revascularization (22 patients), death and nonfatal AMI (18 patients).
- The Global Use of Strategies to Open Occluded Coronary Arteries IV (GUSTO-IV) trial found that elevated troponin levels predicted short-term prognosis regardless of creatinine clearance. Patients with end-stage renal disease (ESRD) with elevated troponin levels had an increased risk of death after 1, 2, and 3 years of follow-up, and increase in troponin levels in ESRD patients showed a two- to fivefold increase in mortality.[53]

2010: highly sensitive cTn

- These are newer high-sensitivity assays that can enable more precise measurement of very low concentrations of cardiac-specific troponin, and experts recommend these to detect cTn in more than 50% of an apparently healthy population.
- Even low-level elevation of troponin detected with these sensitive assays is able to detect cardiac microinfarctions and is associated with worst prognosis.

2001: BNP and NT-proBNP

- These are used to diagnose and grade the severity of congestive heart failure. NT-proBNP is a marker in the blood for BNP, a hormone that rises during cardiac stress due to stretching of chambers of heart.
- These mainly are used to differentiate between heart failure and lung disease (noncardiac) in the patients arriving in emergency with acute onset dyspnea.
- These also guide the monitoring of the effects of therapy for heart failure.

It is considered positive if its value is >100 ng/mL.

Brain natriuretic peptide was prospectively studied as a biomarker of cardiovascular events in CKD patients in a community-based study in Japan.[54] The investigators collected baseline plasma BNP, serum creatinine, and urinary protein levels from 9,625 patients. The risk of cardiovascular events was significantly higher in participants with the highest BNP serum levels. In a study of 134 hemodialysis patients, Sommerer et al[55] found that plasma levels of NT-proBNP were elevated in 100% and cTnT levels in approximately 40% of asymptomatic patients. Both increased NT-proBNP and cTNT were strongly associated with an adverse cardiovascular outcome (overall, there were 23 deaths due to MI and sudden cardiac death).

From recent studies, it has emerged that the long- and short-term prognostic power of BNP and NT-proBNP is similar, in the AMI with ST elevation and without ST elevation, both at hospital admission and during hospitalization.[56]

Brain natriuretic peptide and NT-proBNP have also emerged as prognostic indicators of long-term mortality early after an acute coronary event. This association was observed across the spectrum of ACS, including patients with STEMI, NSTEMI, and unstable angina (UA), those with or without elevated cTn, and those with or without clinical evidence of heart failure.[57,58] Substudies of large-scale clinical trials have evaluated the prognostic value of BNP and NT-proBNP in patients presenting with NSTE-ACS.[59-63] In all studies, elevated values of BNP and NT-proBNP have consistently been found. In a recent analysis made by the group of Sabatine,[60] in 450 patients of the OPUS-TIMI 14 and in 1,635 patients of the TACTICS-TIMI18[57] in which was investigated an approach with multiple markers in ACS without ST elevation, BNP, CRP, and cTnI were all independent predictors of adverse outcome.[60] The incidence of the adverse events not only correlated with the positivity of each marker but also with the number of positive markers, though patients with the worse prognosis (with a relative risk of death to 30 days between 6.0 and 13.0) were those with combined increase of levels of cTnI (marker of thrombosis and myocardial necrosis) of CRP (expression of inflammatory status), and of BNP (marker of myocardial dysfunction). Quite recently, plasma natriuretic peptide concentrations were also related to risk of cardiovascular events and death in apparently asymptomatic persons.[64]

Recent data suggest that natriuretic peptides may help to better identify the very high risk patients and the very low risk ones among a population of ACS patients with respectively, positive or negative troponin. The role of BNP and NT-proBNP testing is included in the

guidelines for the diagnosis and treatment of chronic heart failure of the taskforce of the European Society of Cardiology, published in 2005. Assessment of both markers is considered to be a reliable rule-out test of heart failure in primary care and in the emergency room. Assessment of BNP for risk stratification is also mentioned in the guidelines of the European Society of Cardiology for management of ACS in patients presenting without persistent ST segment elevation, but without any recommendations for the application of BNP and NT-proBNP in clinical routine.[65]

Although originally BNP and NT-proBNP were considered biomarkers for heart failure only, now they are also considered biomarkers of myocardial ischemia. Elevated BNP and NT-proBNP levels have been observed in patients with stable CAD, in patients with UA,[66] and during and after PCI.[67]

RECOMMENDATIONS FOR USE OF BIOCHEMICAL MARKERS FOR DIAGNOSIS OF MI

- Recommended for all patients complaining of chest pain (with clinical examination and ECG)
- Sample timing: on admission
  - 3 to 6 hours later (only when uncertainty exists)
- Test should be with low turnaround time:
  - less than 1 hour (accepted)
  - less than half an hour is preferred
- Types of markers used:

Early markers: as myoglobin and CK-MB; these appear in blood early (within <4 hours) but not specific and does not persist for long period (less than 2 days)

Definitive markers: Troponin—it appears in blood later than myoglobin (within 6 hours) but 100% specific, prognostic, and stays longer (1 week). Its value should be above the 99th percentile of URL.

- Troponin is currently the marker of choice

It should be available in all cardiac and emergency centers (if not, CK-MB mass method for isoforms is the second choice)

EMERGING BIOMARKERS IN ACS (TABLE 2)[68]

Heart-type Fatty Acid-binding Protein (H-FABP)

Heart-type fatty acid-binding protein is a small (15 kDa) soluble nonenzyme protein. It is composed of 132 amino acids. It is one of the most abundant proteins in the heart and comprises 5 to 15% of the total cytosolic protein pool in the aqueous cytoplasm. It was introduced by Glatz et al[69] in 1988 as a potential novel biochemical marker for the early diagnosis of AMI; this was soon confirmed in other studies.[70,71] Under normal conditions, H-FABP is not present in plasma or interstitial fluid, but is released into the blood upon cardiac cellular injury. The cytoplasmic to vascular concentration of H-FABP is of the order of 200,000:1.[72] The plasma or serum concentration of H-FABP under normal conditions is <5 µg/L. This makes the plasma estimation of H-FABP a suitable indicator for the early detection and quantification of myocardial tissue injury.

Górski et al[73] reported that H-FABP and myoglobin concentrations were both significantly elevated in patients with renal failure. The concentrations of these markers were not affected by dialysis. We have also shown that the efficiency of H-FABP for the diagnosis of AMI is severely limited in patients with renal failure.[74]

Table 2: Emerging biomarkers in ACS

Marker name	*Description*
Markers that predict death and/or ischemic event	
Growth differentiation factor 15	Member of TGF-β cytokine family. Released from cardiomyocytes after ischemia and reperfusion injury
Heart-type fatty acid-binding protein	Cytoplasmic protein involved in intracellular uptake and buffering of free fatty acids in myocardium
Myeloperoxidase	Hemeprotein released during degranulation of neutrophils and some monocytes
Pregnancy-associated plasma protein-A	Metaloproteinase expressed in eroded and ruptured plaque but minimally in stable plaque
Placental growth factor	Member of vascular endothelial growth factor family. Strongly upregulated in atherosclerotic plaque instability
Secretory phospholipase A2	Hydrolyzes phospholipids to generate lysophospholipids and fatty acids, thereby enhancing susceptibility of vessels to atherogenesis
Interleukin-6	Stimulator of hepatic synthesis of CRP
Chemokine ligand-5 and ligand-18	Mediators of monocyte recruitment induced by ischemia
Markers that predict heart failure	
Midregional proadrenomedullin	Peptide fragment of vasodilatory peptide adrenomedullin
Neopterin	Marker of monocyte activation
Osteoprotegerin	Modulator of immune function and inflammation

CONCLUSION

The markers that are well suited for the early diagnosis of AMI within the time interval of 0 to 6 hours after symptom onset are myoglobin, H-FABP, and CK-MB isoforms. Although all have been shown to be excellent sensitive early markers, there are still significant issues concerning their specificity. Heart-type fatty acid-binding protein is more cardio-specific than myoglobin and the use of H-FABP as a marker for the early diagnosis of AMI seems preferable. Creatine kinase MB isoenzyme mass measurement is suitable in the 6 to 24 hours interval; CK-MB based on activity measurement is more sensitive in the 12 to 24 hours interval, and the other cardiac markers like total CK, cTnT, and cTnI are most reliable after 12 hours from symptom onset. The prolonged diagnostic window of cTn of several days that is highly sensitive and specific obviates the need for less specific markers with long diagnostic window like AST and CK. Decision-making regarding triage and treatment of patients should not be based on a single measurement of cardiac markers alone because of the time delay required for the marker to exceed the upper limit of reference range. Based on the recent recommendations by the ESC/ACC, cTnI and cTnT are the best markers for the confirmation of AMI. Creatine kinase MB isoenzyme (preferably mass) is the second best marker in the absence of troponin assays. Patients who present with acute chest pain suspicious of ACS are best managed within specifically designated units that have rapid access to specific equipment (ECG, echocardiogram) and facilities (point of care instruments to measure cardiac markers), and with appropriate staffing.

A serial combination testing of a sensitive early marker (e.g., H-FABP, myoglobin, or CK-MB isoforms) and one of the cardiac-specific troponins (cTnT or cTnI) offers the best approach. Two serial testings within a minimum of 12 hours (e.g., at 0, 4–6, or 12 hours) after symptom onset provide reliable sensitivity and specificity for detecting ischemia and evolving infarction within the time interval required for the implementation of reperfusion therapy.

REFERENCES

1. Selker HP, Zalenski RJ, Antman EM, Aufderheide TP, Bernard SA, Bonow RO, Gibler WB, Hagen MD, Johnson P, Lau J, et al. An evaluation of technologies for identifying acute cardiac ischemia in the emergency department: a report from a National Heart Attack Alert Program Working Group. Ann Emerg Med 1997 Jan; 29(1):13-87.
2. Singh TP, Nigam AK, Gupta AK, Singh B. Cardiac biomarkers: when to test? – Physician perspective. J Indian Acad Clin Med 2011;12(2):117-121.
3. Alpert JS, Thygesen K, Antman E, Bassand JP. 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 Sep;36(3):959-969.
4. Fox KA, Eagle KA, Gore JM, Steg PG, Anderson FA; GRACE and GRACE2 Investigators. The Global Registry of Acute Coronary Events, 1999 to 2009-GRACE. Heart 2010 Jul;96(14):1095-1101.
5. Ajay VS, Prabhakaran D. Coronary heart disease in Indians: implications of the INTERHEART study. Indian J Med Res 2010 Nov;132:561-566.
6. Morrow DA, Braunwald E. Future of biomarkers in acute coronary syndromes: moving toward a multimarker strategy. Circulation 2003 Jul;108(3):250-252.
7. Jaffe AS, Babuin L, Apple FS. Biomarkers in acute cardiac disease: the present and the future. J Am Coll Cardiol 2006 Jul;48(1):1.
8. Penttila K, Koukkunen H, Halinen M, Rantanen T, Pyörälä K, Punnonen K, Penttilä I. Myoglobin, creatine kinase MB isoforms and creatine kinase MB mass in early diagnosis of myocardial infarction in patients with acute chest pain. Clin Biochem 2002 Nov;35(8):647.
9. Morrow DA, Antman EM. Evaluation of highly – sensitive assays for cardiac troponin. Clin Chem 2009 Jan;55(1): 53-55.
10. Angurana DK, Lone NA, Khan KA, Jalal S, Sangral R, Rather HA, Alai MS, Habib K, Bhogal BN, Jan VM. Rapid measurement of B- type natriuretic peptide in the diagnosis of congestive heart failure in patients presenting to the emergency department with acute shortness of breath. Int J Med Sci 2011 Mar;3(3):77-82.
11. Ladue JS, Wróblewski F, Karmen A. Serum glutamic oxaloacetic transaminase activity in human acute transmural myocardial infarction. Science 1954 Sep;120(3117):497-499.
12. Karmen A, Wroblewski F, LaDue JS. Transaminase activity in human blood. J Clin Invest 1955 Jan;34(1):126-131.
13. Penttilä I, Penttilä K, Rantanen T. Laboratory diagnosis of patients with acute chest pain. Clin Chem Lab Med 2000 Mar;38(3):187-197.
14. Johnston CC, Bolton EC. Cardiac enzymes. Ann Emerg Med 1982 Jan;11(1):27-35.
15. Wróblewski F, Ross C, Gregory K. Isoenzymes and myocardial infarction. N Engl J Med 1960 Sep;263:531-536.
16. Konttinen A. Hydroxybutyrate dehydrogenase in detection of myocardial infarction. Lancet 1961 Sep;2(7201):556-558.
17. Galbraith LV, Leung FY, Jablonsky G, Henderson AR. Time related changes in the diagnostic utility of total lactate dehydrogenase, lactate dehydrogenase isoenzyme-1, and two lactate dehydrogenase isoenzyme-1 ratios in serum after myocardial infarction. Clin Chem 1990 Jul;36(7):1317-1322.
18. Hill BR, Levi C. Elevation of a serum component in neoplastic disease. Cancer Res 1954 Aug;14(7):513-515.
19. Wroblewski F, LaDue JS. Lactic dehydrogenase activity in blood. Proc Soc Exp Biol Med 1955 Oct;90(1):210-213.
20. Wróblewski F, Ruegsegger P, LaDue JS. Serum lactic dehydrogenase activity in acute transmural myocardial infarction. Science 1956 Jun;123(3208):1122-1123.
21. Ulmer DD, Vallee BL, Wacker WE. Metalloenzymes and myocardial infarction. II. Malic and lactic dehydrogenase activities and zinc concentrations in serum. N Engl J Med 1956 Sep;255(10):450-456.
22. Dreyfus JC, Schapira G, Rasnais J, Scebat L. Serum creatine kinase in the diagnosis of myocardial infarct. Rev Fr Etud Clin Biol 1960 Apr;5:386-387.

23. Sorensen NS. Creatine phosphokinase in the diagnosis of myocardial infarction. Acta Med Scand 1963 Jan/Dec;174(6): 725-734.
24. Shell WE, Kjekshus JK, Sobel BE. Quantitative assessment of the extent of myocardial infarction in the conscious dog by means of analysis of serial changes in serum creatine phosphokinase activity. J Clin Invest 1971 Dec;50(12):2614-2625.
25. Sobel BE, Bresnahan GF, Shell WE, Yoder RD. Estimation of infarct size in man and its relation to prognosis. Circulation 1972 Oct;46(4):640-648.
26. Roe CR, Limbird LE, Wagner GS, Nerenberg ST. Combined isoenzyme analysis in the diagnosis of myocardial injury: Application of electrophoretic methods for the detection and quantitation of the creatine phosphokinase MB isoenzyme. J Lab Clin Med 1972 Oct;80(4):577-590.
27. Panteghini M. Enzyme and muscle diseases. Curr Opin Rheumatol 1995;7:469-474.
28. Puleo PR, Guadagno PA, Roberts R, Scheel MV, Marian AJ, Churchill D, Perryman MB. Early diagnosis of acute myocardial infarction based on assay for subforms of creatine kinase-MB. Circulation 1990 Sep;82(3):759-764.
29. Wu AH, Wang XM, Gornet TG, Ordóñez-Llanos J. Creatine kinase MB isoforms in patients with skeletal muscle injury: ramifications for early detection of acute myocardial infarction. Clin Chem 1992 Dec;38(12):2396-2400.
30. Zimmerman J, From R, Meyer D, Boudreaux A, Wun CC, Smalling R, Davis B, Habib G, Roberts R. Diagnostic marker cooperative study for the diagnosis of myocardial infarction. Circulation 1999 Apr;99(13):1671-1677.
31. Seo H, Miyazaki S, Furuno T, Nonogi H, Haze K, Hiramori K. Creatine kinase-MB protein mass is a better indicator for the assessment of acute myocardial infarction in the lower range of creatine kinase level. Jpn Heart J 1993 Nov;34(6): 717-727.
32. Pettersson T, Ohlsson O, Tryding N. Increased CK-MB (mass concentration) in patients without traditional evidence of acute myocardial infarction. A risk indicator of coronary death. Eur Heart J 1992 Oct;13(10):1387-1392.
33. Ravkilde J, Nissen H, Mickley H, Andersen PE, Thayssen P, Horder M. Cardiac troponin-T and CK-MB mass release after visually successful percutaneous transluminal coronary angioplasty in stable angina pectoris. Am Heart J 1994;127: 13-20.
34. Abbas SA, Glazier JJ, Wu AH, Dupont C, Green SF, Pearsall LA, Waters DD, McKay RG. Factors associated with the release of cardiac troponin T following percutaneous transluminal coronary angioplasty. Clin Cardiol 1996 Oct;19(10):782-786.
35. Pearson JR, Carrea F. Evaluation of the clinical usefulness of a chemiluminometric method for measuring creatine kinase. Clin Chem 1990.;36:1809.
36. Gilkeson G, Stone MJ, Waterman M, Ting R, Gomez-Sanchez CE, Hull A, Willerson JT. Detection of myoglobin by radioimmunoassay in human sera: its usefulness and limitations as an emergency room screening test for acute myocardial infarction. Am Heart J 1978 Jan;95(1):70-77.
37. Varki AP, Roby DS, Watts H, Zatuchni J. Serum myoglobin in acute myocardial infarction: a clinical study and review of the literature. Am Heart J 1978 Nov;96(5):680-688.
38. Bhayana V, Cohoe S, Leung FY, Jablonsky G, Henderson AR. Diagnostic evaluation of creatine kinase-2 mass and creatine kinase-3 and -2 isoform ratios in early diagnosis of acute myocardial infarction. Clin Chem 1993;39:488-495.
39. Greaser ML, Gergely J. Reconstitution of troponin activity from three protein components. J Biol Chem 1971;246: 4226-4233.
40. Katus HA, Remppis A, Looser S, Hallermeier K, Scheffold T, Kübler W. Enzyme linked immuno assay of cardiac troponin T for the detection of acute myocardial infarction in patients. J Mol Cell Cardiol 1989 Dec;21(12):1349-1353.
41. Horwich TB, Patel J, MacLellan WR, Fonarow GC. Cardiac troponin I is associated with impaired hemodynamics, progressive left ventricular dysfunction, and increased mortality rates in advanced heart failure. Circulation 2003 Aug 19;108(7):833-838.
42. Ni CY. Cardiac troponin I: a biomarker for detection and risk stratification of minor myocardial damage. Clin Lab 2001; 47(9-10):483-492.
43. Ohman EM, Armstrong PW, Christenson RH, Granger CB, Katus HA, Hamm CW, O'Hanesian MA, Wagner GS, Kleiman NS, Harrell FE Jr, et al. Cardiac troponin T levels for risk stratification in acute myocardial ischemia. GUSTO IIA Investigators. N Engl J Med 1996 Oct 31;335(18):1333-1341.
44. Apple FS, Murakami MM, Pearce LA, Herzog CA. Predictive value of cardiac troponin I and T for subsequent death in end-stage renal disease. Circulation 2002 Dec 3;106(23): 2941-2945.
45. Evaluation of a bedside whole-blood rapid troponin T assay in the emergency department. Rapid Evaluation by Assay of Cardiac Troponin T (REACTT) Investigators Study Group. Acad Emerg Med 1997 Nov;4(11):1018-1024.
46. Hamm CW, Goldmann BU, Heeschen C, Kreymann G, Berger J, Meinertz T. Emergency room triage of patients with acute chest pain by means of rapid testing for cardiac troponin T or troponin I. N Engl J Med 1997 Dec 4;337(23):1648-1653.
47. Li D, Jialal I, Keffer J. Greater frequency of increased cardiac troponin T than increased cardiac troponin I in patients with chronic renal failure. Clin Chem 1996 Jan;42(1):114-115.
48. Needham DM, Shufelt KA, Tomlinson G, Scholey JW, Newton GE. Troponin I and T levels in renal failure patients without acute coronary syndrome: a systematic review of the literature. Can J Cardiol 2004 Oct;20(12):1212-1218.
49. Khan NA, Hemmelgarn BR, Tonelli M, Thompson CR, Levin A. Prognostic value of troponin T and I among asymptomatic patients with end-stage renal disease: a meta-analysis. Circulation 2005 Nov 15;112(20):3088-3096.
50. Jurlander B, Farhi ER, Banas JJ Jr, Keany CM, Balu D, Grande P, Ellis AK. Coronary angiographic findings and troponin T in patients with unstable angina pectoris. Am J Cardiol 2000 Apr 1;85(7):810-814.
51. Lindahl B, Venge P, Wallentin L. Relation between troponin T and the risk of subsequent cardiac events in unstable coronary artery disease. The FRISC study group. Circulation 1996 May 1;93(9):1651-1657.
52. Stubbs P, Collinson P, Moseley D, Greenwood T, Noble M. Prospective study of the role of cardiac troponin T in patients admitted with unstable angina. BMJ 1996 Aug 3;313(7052): 262-264.
53. Aviles RJ, Askari AT, Lindahl B, Wallentin L, Jia G, Ohman EM. Troponin T levels in patients with acute coronary syndromes, with or without renal dysfunction. N Engl J Med 2002;346: 204-205.
54. Sakuma M, Nakamura M, Tanaka F, Onoda T, Itai K, Tanno K, Ohsawa M, Sakata K, Yoshida Y, Kawamura K, et al. Plasma B-type natriuretic peptide level and cardiovascular events in

chronic kidney disease in a community-based population. Circ J 2010;74(4):792-797.
55. Sommerer C, Beimler J, Schwenger V, Heckele N, Katus HA, Giannitsis E, Zeier M. Cardiac biomarkers and survival in haemodialysis patients. Eur J Clin Invest 2007;37(5):350-356.
56. Galvani M, Ottani F, Oltrona L, Ardissino D, Gensini GF, Maggioni AP, Mannucci PM, Mininni N, Prando MD, Tubaro M, et al. N-terminal pro-brain natriuretic peptide on admission has prognostic value across the whole spectrum of acute coronary syndromes. Circulation 2004 Jul 13;110(2): 128-134.
57. de Lemos JA, Morrow DA. Brain natriuretic peptide measurement in acute coronary syndromes: ready for clinical application? Circulation 2002 Dec 3;106(23):2868-2870.
58. White HD, French JK. Use of brain natriuretic peptide levels for risk assessment in non-ST-elevation acute coronary syndromes. J Am Coll Cardiol. 2003 Dec 3;42(11):1917-1920.
59. Varo N, de Lemos JA, Libby P, Morrow DA, Murphy SA, Nuzzo R, Gibson CM, Cannon CP, Braunwald E, Schönbeck U. Risk prediction after acute coronary syndromes. Circulation 2003 Sep 2;108(9):1049-1052.
60. Sabatine MS, Morrow DA, de Lemos JA, Gibson CM, Murphy SA, Rifai N, McCabe C, Antman EM, Cannon CP, Braunwald E. Multimarker approach to risk stratification in non-ST elevation acute coronary syndromes: simultaneous assessment of troponin I, C-reactive protein, and B-type natriuretic peptide. Circulation 2002 Apr 16;105(15):1760-1763.
61. Lindahl B, Lindbäck J, Jernberg T, Johnston N, Stridsberg M, Venge P, Wallentin L. Serial analyses of N-terminal pro-B-type natriuretic peptide in patients with non-ST-segment elevation acute coronary syndromes: a Fragmin and fast Revascularisation during In Stability in Coronary artery disease (FRISC)-II substudy. J Am Coll Cardiol 2005 Feb 15;45(4):533-541.
62. Jernberg T, James S, Lindahl B, Stridsberg M, Venge P, Wallentin L. NT-proBNP in unstable coronary artery disease–experiences from the FAST, GUSTO IV and FRISC II trials. Eur J Heart Fail 2004 Mar 15;6(3):319-325.
63. Heeschen C, Dimmeler S, Fichtlscherer S, Hamm CW, Berger J, Simoons ML, Zeiher AM, CAPTURE Investigators. Prognostic value of placental growth factor in patients with acute chest pain. JAMA 2004 Jan 28;291(4):435-441.
64. Wang TJ, Larson MG, Levy D, Benjamin EJ, Leip EP, Omland T, Wolf PA, Vasan RS. Plasma natriuretic peptide levels and the risk of cardiovascular events and death. N Engl J Med 2004 Feb 12; 350(7):655-663.
65. Kay JD, Trichon BH, Kisslo M, Pentagini M. Serum brain natriuretic peptide levels cannot differentiate pulmonary disease from left-heart failure if the right ventricle is dilated. Circulation 2003;108 IV:397.
66. Kikuta K, Yasue H, Yoshimura M, Morita E, Sumida H, Kato H, Kugiyama K, Ogawa H, Okumura K, Ogawa Y, et al. Increased plasma levels of B-type natriuretic peptide in patients with unstable angina. Am Heart J 1996 Jul;132(1 Pt 1):101-107.
67. Tateishi J, Masutani M, Ohyanagi M, Iwasaki T. Transient increase in plasma brain (B-type) natriuretic peptide after percutaneous transluminal coronary angioplasty. Clin Cardiol. 2000 Oct;23(10):776-780.
68. Kempf T, Eden M, Strelau J, Naguib M, Willenbockel C, Tongers J, Heineke J, Kotlarz D, Xu J, Molkentin JD, et al. The transforming growth factor – Beta superfamily member growth – differentiationfactor – 15 protects the heart from ischemia. Circ Res 2006;98(3):351-360.
69. Glatz JF, van Bilsen M, Paulussen RJ, Veerkamp JH, van der Vusse GJ, Reneman RS. Release of fatty acid-binding protein from isolated rat heart subjected to ischemia and reperfusion or to the calcium paradox. Biochim Biophys Acta 1988 Jul 1;961(1):148-152.
70. Ishii J, Wang JH, Naruse H, Taga S, Kinoshita M, Kurokawa H, Iwase M, Kondo T, Nomura M, Nagamura Y, et al. Serum concentrations of myoglobin vs human heart-type cytoplasmic fatty acid-binding protein in early detection of acute myocardial infarction. Clin Chem 1997 Aug;43(8 Pt 1):1372-1378.
71. Alhadi HA, Fox KA. Do we need additional markers of myocyte necrosis: the potential value of heart fatty-acid-binding protein. QJM 2004 Apr;97(4):187-198.
72. Glatz JF, Van der Vusse GJ, Maessen JG, Van Dieijen-Visser MP, Hermens WT. Fatty acid-binding protein as marker of muscle injury: experimental findings and clinical application. Acta Anaesthesiol Scand Suppl 1997;111:292-294.
73. Górski J, Hermens WT, Borawski J, Mysliwiec M, Glatz JF. Increased fatty acid-binding protein concentration in plasma of patients with chronic renal failure. Clin Chem 1997 Jan;43(1): 193-195.
74. Al-Hadi HA, William B, Fox KA. Serum level of heart-type Fatty Acid-binding protein in patients with chronic renal failure. Sultan Qaboos Univ Med J 2009 Dec;9(3):311-314.