Original Research Article

The utility of Heart type Fatty Acid Binding Protein in Acute Myocardial Infarction Patients

Subrat Chandra[1], Manish Raj Kulshrestha[2], Apurva Raj[3*], Bhuwan Chandra Tiwari[4]

[1]Department of Pathology, Dr. Ram Manohar Lohia Institute of Medical Sciences, Lucknow Uttar Pradesh India
[2]Department of Biochemistry, Dr. Ram Manohar Lohia Institute of Medical Sciences, Lucknow Uttar Pradesh India
[3]Department of Pathology, Superspeciality Cancer Institute, Lucknow Uttar Pradesh India
[4]Department of Cardiology, Dr. Ram Manohar Lohia Institute of Medical Sciences, Lucknow Uttar Pradesh India

Abstract

Introduction: Early diagnosis and therapeutic intervention can improve the outcome of acute myocardial infarction (AMI). However, there are no satisfactory cardiac biomarkers for the diagnosis of AMI within 6 hours of onset of symptoms. Among the novel biomarkers of AMI, we tested the diagnostic accuracy of Heart type Fatty Acid Binding Protein (HFABP) in prospectively recruited patients with independently adjudicated outcomes. *Methods:* Prospective observational study was conducted at the Department of Pathology and Department of Cardiology, Dr Ram Manohar Lohia Institute of Medical Sciences Lucknow. After taking informed consent, 80 cases of chest pain suggestive of coronary origin (within 6 hours) were included. *Results:* Group a included 55 patients while Group B included 25 patients. In our study on Receiver operator curve (ROC) analysis, the AUC (Area under curve) was 0.846 (95% C.I. 0.743 to 0.948; p value <0.001) for HFABP. The optimum cut-off value for HFABP to diagnose AMI was 6.38 ng/ml with a Sensitivity, Specificity, PPV (Positive Predictive Value) and NPV of 86.4%, 84.5%, 67.9% and 94.2% respectively. *Conclusion:* The optimum cut-off value for HFABP to diagnose AMI was found to be 6.38 ng/ml were in concordance with similar studies. A high sensitivity and NPV is essential for the early 'rule out' of AMI patients. Since more than 70% of patients who present with acute chest pain to an emergency department do not have AMI, HFABP can be used as rule out tool to prevent unnecessary admissions of patients suspected for AMI considering its high sensitivity and NPV. Though, it was not a reliable marker for AMI diagnosis because of low specificity and positive predictive value. Further larger studies are needed to understand whether HFABP can add incremental value in rule-in AMI.
Keywords: Acute myocardial infarction, Heart type fatty acid binding protein, Rule out Tool.

INTRODUCTION

Myocardial infarction is defined by the clinical history, electrocardiogram, and an increase or decrease in cardiac troponin concentration (as evidence of myocardial necrosis) [1]. Assessing chest pain patients presenting to the emergency area (EA) is still a clinical challenge, as acute myocardial infarction (AMI) diagnosis is not adjudicated in the majority of patients. Acute Myocardial Infarction (AMI) accounts for 5-15% of all the causes of chest pain in USA. In countries like India, considerably higher number of patients present with chest pain as the chief complaint to the emergency department. The incidence of MI in India is 64.37/1000 people [2]. The overall prevalence of coronary heart disease (CHD) in Lucknow district was found to be 7.1% with 8.8% in urban areas and 3.8% in rural areas [3]. Epidemiological studies showed 29.8 million coronary heart disease patients in India and 1.27 million

acute coronary events resulting in 20,000 coronary bypass surgeries and 1,113,359 coronary angioplasties per year. As per World Health Organisation (WHO) data, the Coronary Artery Disease (CAD) prevalence continues to rise in India with rapid 'epidemiological transition'. It has already surpassed communicable diseases as the major cause of mortality in India. It has been projected that between 1990 and 2020, there will be 117% and 105% rise in mortality from CAD in men and women respectively in India [4]. In a typical population of patients undergoing evaluation for acute chest pain in emergency department, about 15% to 25% have AMI. By accurately ruling out chest pain of cardiac origin, 40% of patients presenting with acute chest pain could be spared from the risks and costs of unnecessary hospital admission and more invasive cardiac testing [2]. A missed diagnosis of acute coronary syndrome (ACS) may lead to further ischemic

events in a potentially preventable death or disability. Therefore, patients with symptoms suggestive of ACS often undergo a lengthy assessment in the emergency department (ED) or as hospital in patients. These patients account for approximately 10% of Emergency Department presentations and 25% of hospital admissions, yet up to 85% do not have a final diagnosis of ACS. International guidelines for the investigation of ACS recommend serial measurement of cardiac troponin (cTn) at the onset of symptoms which is more cumbersome process for identifying patients with low short-term risk of adverse cardiac events and to support their earlier discharge.

Among all, Heart type fatty acid binding protein (HFABP) has been largely investigated. It is a small (15kDa) soluble protein, present in cardiomyocytes cytoplasm at high concentrations [5]. It is rapidly released into plasma after the onset of myocardial injury, with a peak at approximately 6-8 hours [6].

METHODS

Prospective observational study was conducted at the Department of Pathology and Department of Cardiology, Dr Ram Manohar Lohia Institute of Medical Sciences Lucknow, from February 2016 to September 2018 after institutional ethical clearance (IEC number 31/16). The study included 80 cases of Chest pain suggestive of coronary origin on the basis discretion of emergency department cardiologist assessment and/or ECG findings.

The patients admitted to cardiology department at RMLIMS fulfilling inclusion criteria will be taken in study. 2 ml blood sample in EDTA vial was taken. HFABP was estimated after centrifugation at 3000 rpm for 10 minutes, the serum was separated. Following serum creatinine was estimated by modified Jaffe's assay kinetic assay on Beckmann coulter AU480. HFABP was estimated by Enzyme Linked Immune Sorbent Assay (ELISA), based on biotin double antibody sandwich technology respectively (Biovendor research and diagnostic products).

Groups were formed and compared: The patients was further subgrouped based on the basis of onset of chest pain-

Group A: Time of first sampling is within 3 hours of onset of chest pain

Group B: Time of first sampling is >3 hours of onset of chest pain

The performance of HFABP was analysed with their serum levels in sample taken at the time of presentation in terms of sensitivity, specificity, positive predictive value and negative predictive value. All these

performances were analysed and compared with SPPSS version 21.

Patients presenting with acute chest pain of less than six-hour duration suspected of coronary origin were enrolled. These patients were also randomly classified according time of chest pain onset into two groups (<3 hours and >3 hours). Full history taking including patients' symptoms and past medical history such as diabetes mellitus (DM), hypertension (HTN) and previous ischemic event; and general clinical examination, ECG and laboratory investigations were documented at admission using a pre-defined protocol.

The standard diagnosis was made after critical review of all the clinical pictures and relevant information by a senior cardiologist. AMI was defined according to the European Society of Cardiology/American College of Cardiology Committee Criteria. AMI was defined as detection of initial or 0-3 h hsTnI above 99 percentile upper reference limit (URL) together with evidence of myocardial ischemia with at least one of the following:

(i). Symptoms of ischemia
(ii). ECG changes indicative of new ischemia (new ST-T changes or new left bundle branch block [LBBB]
(iii).Development of pathological Q waves in the ECG
(iv). Imaging evidence of new loss of viable myocardium or new regional wall motion abnormality.

Diagnostic outcomes were first categorized into the following groups:
(i).ST-elevation myocardial infarction (STEMI)
(ii). Non-ST-elevation myocardial infarction (NSTEMI)
(iii). Unstable angina (UA)
(iv). Non-cardiac chest pain (NCCP).

The first two groups were later integrated into AMI group, and the last two into non-myocardial infarction (non-AMI) group.

The diagnosis of chest pain cases as myocardial infarction will be taken as finalized by cardiologists.

INCLUSION CRITERIA
1. Age > 18 Years
2. Consent taken.
3. Chest pain suggestive of coronary origin on the basis discretion of emergency physician/ cardiologist assessment and/or ECG findings with with at least one hsTnI level above the upper 99th percentile (Upper Reference Limit or URL).
Chest pain suggestive of coronary origin is defined, in accordance with ACC/AHA guidelines, as chest or left arm pain as the chief symptom. ECG changes suggestive of ischemia are defined as pathologic Q waves of more than 40msec, ST-

segment elevation or depression of more than 1mm or abnormal T wave morphology.

EXCLUSION CRITERIA

1. Chronic renal failure (eGFR below 60 ml/min)
2. Acute history of muscle injury/ trauma
3. Patients not willing to provide blood sample
4. Patients with previous history of AMI

Two venous blood samples (2 ml) were withdrawn and blood serum was separated from the clot and aliquoted under complete aseptic condition. The serum obtained was kept frozen at -40 degree centigrade till the analysis of serum creatinine and HFABP.

The H-FABP was determined by enzyme linked immunosorbant assay (ELISA, Biovendor research and diagnostic products). The assay system uses a double antibody sandwich enzyme linked immunosorbent one step process assay (ELISA) to determine the level of HFABP in samples.

RESULTS

In present study eighty consecutive patients with complain of acute chest pain of less than six hours suggestive of cardiac origin were evaluated in the department of cardiology, RMLIMS, Lucknow. Majority of patients belonged to the age group >61 years (40%) and the mean age of presentation was 58.5 Yrs and the main bulk of the study population between 40 to 70 years of age. Incidence of AMI was more common in male, patients with past medical history of hypertension, smoking and diabetes mellitus. In cases diagnosed as AMI, 50% of the patients found to have ST elevation.17.5% of patients were diagnosed as unstable angina and 55% cases were diagnosed as having non-cardiac chest pain. In sub group analysis group, A included 55 patients while sub group B included 25 patients. In our study on Receiver operator curve (ROC) analysis, the AUC (Area under curve) was 0.846 (95% C.I. 0.743 to 0.948; p value <0.001) for HFABP. The optimum cut-off value for HFABP to diagnose AMI was 6.38 ng/ml with a Sensitivity, Specificity, PPV and NPV of 86.4%, 84.5%, 67.9% and 94.2% respectively.
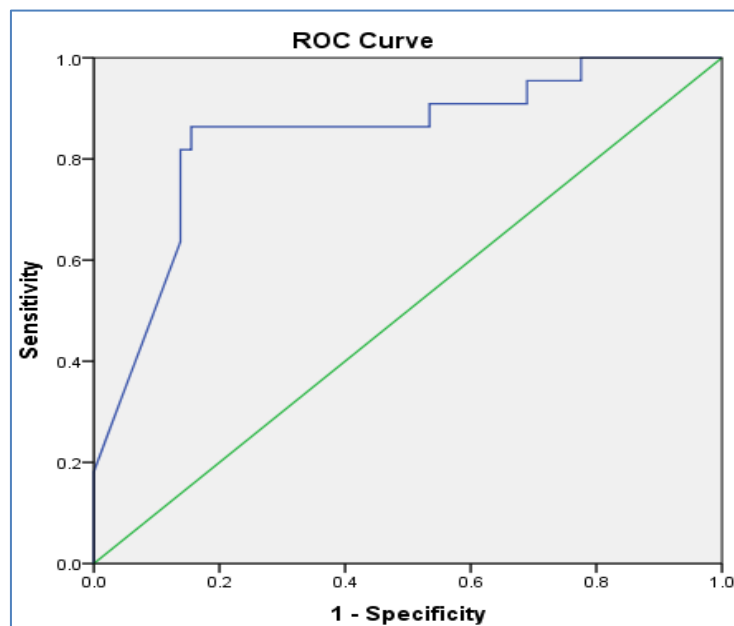
Diagram-1: The ROC analysis of HFABP at the time of admission

Table-1: Analysis of AMI patients using HFABP

TIME GROUP	HFABP ELEVATION	MYOCARDIAL INFARCTION		TOTAL
		Yes	No	
GROUP A Presentation<3hr	Yes	9	6	15
	No	3	37	40
		12	43	55
GROUP B Presentation >3 hr	Yes	10	3	13
	No	0	12	12
		10	15	25

HFABP in group A showed Sensitivity, Specificity, PPV and NPV of 75%,86%, 60% and 92.5% while in group B showed Sensitivity, Specificity, PPV and NPV of 100%,80%,76.9% and 100%.

Table-2: The performance of HFABP (HFABP > 6.38 ng/mL were considered for AMI) at the time of admission

	<3 hours presentation (n=55)	>3 hours presentation (n=25)	Overall
Sensitivity (%)	75	100	86.4
Specificity (%)	86	80	84.5
PPV (%)	60	76.9	67.9
NPV (%)	92.5	100	94.2

In 9 subjects (11.25%) out of 80 acute chest pain patients HFABP was falsely elevated. The most common cause was being unstable angina in our study.

Table-3: High HFABP levels (>6.38 ng/ml) in different subgroups based on final diagnosis

Elevated HFABP	Final diagnosis	n	HFABP >6.38 ng/ml
True Positive (n=19)	STEMI	11	11 (100%)
	NSTEMI	11	08 (72.7%)
False Positive (n=09)	Unstable Angina	14	05 (35.7%)
	Non cardiac chest pain	44	04 (9.1%)
	Total	80	28 (35%)

DISCUSSION

In our study eighty consecutive patients with complain of acute chest pain of less than six hours and suggestive of cardiac origin were evaluated in the department of cardiology, RMLIMS, Lucknow. Incidence of AMI was more common in male, patients with past medical history of hypertension, smoking and diabetes mellitus. The reason being the above-mentioned factors are the known risk factors for coronary heart disease. In our study, we did also find that mean age of presentation was 58 years with 40% patients were above 61 years of age. Majority (86.3%) were males with diabetes mellitus and hypertension significantly more in AMI cases than non AMI patients (p value 0.027 and 0.031 respectively).

In cases diagnosed as AMI, 50% of the patients found to have ST elevation.17.5% of patients were diagnosed as unstable angina and 55% cases were diagnosed as having noncardiac chest pain and out of which 13.7% cases showed T wave inversion. The diagnostic value of the admission ECG is limited if i) the presence of conduction disorders including left bundle branch block (LBBB); ii) If Q waves and ST-T changes are already present e.g. old infarcts. iii) ST-T wave changes of marked left ventricular hypertrophy; iv) In posterior infarct or right ventricular infarct. 30% of patients may have no diagnostic changes on their admission ECG.

In our study on Receiver operator curve (ROC) analysis, the AUC (Area under curve) was 0.846 (95% C.I. 0.743 to 0.948; p value <0.001).

Table-4

Study	Area under Curve (95% Confidence Interval) AUC (95% C.I.)
Kellens S *et al*. [7]	0.830 (0.770–0.890)
Ruff TC *et al*. [8]	0.780 (0.720-0.840)
Eggers KM *et al*. [9]	0.800 (0.760-0.840)
Reddy LL *et al*. [10]	0.728(0.622-0.817)
Present Study	0.846 (0.743 to 0.948)

The optimum cut-off value for HFABP to diagnose AMI was 6.38 ng/ml with sensitivity, specificity, PPV and NPV of 86.4%, 84.5%, 67.9% and 94.2% respectively. The cut off value in our study is in concordance with similar studies.

Table-5: illustrated that the prespecified cut-off value for HFABP varied between studies, a cut-off between5 to 7ng/ml was used, four studies used the same assay and the same cut-off point (>6.38 ng/mL) for abnormality [11]

Author	Study type	Study population	Index test threshold (ng/ml)	Index test assay	Time from symptoms
Vupputuri2015	India, singlecentre	54	6.4	abbott Architect	Up to 6 hours
Pyati 2015	India, Single centre	80	6.3	Roche C-311	<3 Hours
Gerede 2015	Turkey, Single Centre	48	7.0	CardioDetect	<3 Hours
Willemsen 2015	Netherland, Single Centre	88	5.3	Fab Pulous BV	<3 Hours
Gami 2015	India, Single Centre	88	5.1	ELISA	Up to 6 hours
Kellens S 2015	Belgium, Single Centre	203	5.3	Fab Pulous BV	Up to 6 hours
Dupuy AM 2015	France, Single Centre	181	5.8	Randox-Cobas	Up to 6 hours
Kitamura 2013	Japan, Single Centre	63	6.2	ELISA	<3 Hours
Kagawa Y 2013	Japan, Single Centre	114	5.8	ELISA	Up to 6 hours
Garcia 2011	Spain, Single Centre	165	6.2	ELISA	<3 Hours
McMahon 2012	Ireland, Single Centre	1128	5.2	Randox-Cobas	<3 Hours
Xu Q 2010	China, Single Centre	87	7.0	CardioDetect	<3 Hours
Liao J 2009	China, Single Centre	74	7.0	CardioDetect	<3 Hours
Mad P 2007	Austria, Single Centre	107	7.0	CardioDetect	<3 Hours
Seino Y 2004	Japan, Single Centre	35	6.2	ELISA	<3 Hours
Seino Y 2003	Japan, Single Centre	371	6.2	ELISA	<3 Hours
Body R 2011	UK, Single Centre	705	6.8	ELISA	<3 Hours
Reiter M 2013	Switzerland, Single Centre	1074	4.2	QuickSensh	<6 Hours
Ruff CT 2013	US, Single Centre	343	5.0	Randox	Up to 6 hours
Aldous 2012	New Zealand, Single Centre	384	6.0	Hycult	<3 Hours
Eggers 2012	Sweden, Multi Centre	360	5.8	Randox	Up to 6 hours
Collinson 2014	UK, Multi Centre	850	6.0	ELISA	<3 Hours
Reddy LL *et al*.	Chennai, India	88	6.32	Randox	<6 hours
Present Study	Lucknow, Single Center	80	6.3	ELISA	<6Hours

Sensitivity and specificity of H-FABP was significantly high in present study which is consistent with various other studies.

Table-6: The performance of HFABP in different studies

Study	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
Ruff TC *et al*. [8]	63	79	51	85
Van Hise CB *et al*. [12]	99.1	43.4	12.9	100
Fissha MZ *et al*. [13]	75	83.7	30	93.3
Kellens S *et al*. [7]	54	84	85	52
Elma bouh I *et al.* [14]	81.8	88.2	81.8	88.2
Vupputuri *et al*. [15]	100	85.7	88.2	100
Present study	86.4	84.5	67.9	94.2

CONCLUSION

A high sensitivity and NPV is essential for the early 'rule out' of AMI patients. Since more than 70% of patients who present with acute chest pain to an emergency department do not have AMI, H-FABP can be used as rule out tool to prevent unnecessary admissions of patients suspected for AMI considering its high sensitivity and NPV. On the other side it can be stated that HFABP is clearly not a reliable marker for AMI diagnosis because of low specificity and PPV. Relatively larger studies and more interventional trials are needed to understand whether HFABP can add incremental value in rule-in AMI.

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