A SERIAL FOLLOW UP STUDY OF CARDIAC MARKER ENZYMES DURING THE WEEK AFTER ACUTE MYOCARDIAL INFARCTION.

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ABSTRACT

Laboratory infarction diagnostics are based on the detection of elevated serum activities of creatine kinase (CK) Creatine kinase Isoenzyme MB (CKMB) and Transaminases. Determination of these cardiac marker enzymes permits the diagnosis of transmural myocardial infarction. However in such patients the diagnosis of acute myocardial infarction can be confirmed by the clinical symptoms and changes in the ECG, in addition to the enzyme assays. The 50 AMI patients selected in the present study were those admitted to the ICCU of Shri Krishna Hospital, Karamsad. The blood samples were taken at Zero hours (i.e. at the time of admission of the patient). Within 6 hrs of the starting of chest pain, 1.5 million units of streptokinase were mixed with 100 to 150ml of normal saline and administered by infusion over a period of one hour. The blood samples were further collected at intervals of 6 hrs, 14hrs, 32hrs, 48hrs, 5th day and 7th day. The blood samples were analyzed for CK, CKMB, SGOT, α HBDH and Cardiac specific Troponin T. By 6hrs the CK and CKMB values had started rising, the rise continuing at 14hrs with peak values at 32hrs. The CK showed a slight decrease by 48 hrs. The cardiac Troponin T showed wide time window from 4 hrs to 7th day for detecting myocardial damage. The maximum cardiac Troponin T values were during the first 24hrs. Cardiac Troponin T in serum appears to be a more sensitive and early indicator of myocardial cell injury in comparison to CKMB.

KEY WORDS

Cardiac Troponin T, α -Hydroxy butyrate dehydrogenase, Myocardial infarction.

INTRODUCTION

The available parameters are found wanting in patients of multiorgan disease or with additional skeletal muscle lesions, because in such cases a definite differentiation of skeletal muscle and heart muscle necrosis is impossible because of the organ distribution of CK, LDH or their isoforms.

In addition to helping in the diagnosis of acute myocardial infarction (AMI) the laboratory parameters are of major importance in monitoring the course of infarction and in

estimating its size. Cytosol proteins such as CK are washed out in different percentage in dependence on the blood flow in the early phase of the infarction and therefore appear earlier in the reperfused infarct and their plasma activity is higher than non-reperfused infarcts. Although with certain limitations, the difference in the release kinetics of CK can be used to evaluate the outcome of thrombolytic treatment, the perfusion dependent washout of cytosol proteins, once the infarct reperfusion has occurred. It further allows to estimate the size of the infarct by an analysis of CK.

A new immunoassay for Cardiac specific Troponin T (CTnT) that eliminates certain shortcomings of laboratory infarction diagnostics is envisaged to be carried out. The present study is expected to document the value of the new enzyme immunoassay of CTnT. It is important to note that sub acute and silent infarctions in particular can be diagnosed with a degree of reliability. The changes in the serum concentrations of CTnT are reported to differ significantly from the kinetics of enzymes used previously in infarction diagnostics. CTnT

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exhibits serum concentration changes similar to those observed for proteins dissolved in the cytosol (e.g. Myoglobin, CK) and those seen for structurally bound proteins such as light myosin chains. The CTnT determination is likely to become extremely important in monitoring the course and therapy of a myocardial infarction. This applies particularly to follow the outcome of a thrombolytic treatment on the basis of the course kinetics of the serum CTnT level and hence this study.

MATERIALS AND METHODS

The study carried out incorporates the serial estimation of different cardiac markers in-patients having myocardial infarction. They were admitted to the ICCU of Shri Krishna Hospital, Karamsad. The first sample was collected at the time of admission of the patient. Within 6hrs of the starting of chest pain 1.5 million units of streptokinase mixed with 100 to 150 ml of normal saline was administered by intravenous infusion over a period of one hour. The blood samples were collected at intervals of 6hrs, 32hrs, 48hrs, further 5th day and 7th day. The samples were collected in plain vials and were allowed to clot and serum separated. The samples were analyzed for CK and CKMB on the same day. The SGOT and α HBDH were estimated within two days. For Troponin T estimation the samples were kept at -20°C till the analysis. ie. Within 7 days. Care was taken that the samples were not hemolyzed. The samples were collected by using disposable needle and syringe.

Sample Collection : From the samples collected in plain vials at different time intervals as mentioned above, the serum was separated by centrifugation at 5000 rpm. These sera were used for the estimation of CK, CKMB, SGOT, α HBDH and Troponin T on RA -50 semi auto analyzer by well standardized methods (1-5). The CTnT was estimated by the reagent kit manufactured by Boehringer based on ELISA. The study comprised of 50 patient with AMI out of which 12 were females and 38 were males. Twenty-five subjects were selected from normal blood donors and another 25 subjects with

polytraumatic injury without any chest or cardiac ailment were also included in the study as controls.

RESULTS AND DISCUSSION

Myocardial infarction can occur at any age irrespective of sex but generally it is a disease of middle and old age. 44% in our study belonged to the age group of 60-70 years, 30% were in the range of 50-59 years and 26% had an age between 40 and 49.

A number of studies have been carried out to show the utility of ECG changes in the acute myocardial infarction. Pardec (1920) was the first to observe the ST segment changes in coronary artery disease. He also observed an alteration in ECG in the course of myocardial infarction. These changes also have been observed in the absence of anatomical evidence of myocardial damage. The report of other workers like Bhimgart et al (1937) did not find ST segment changes in the presence of significant myocardial necrosis. (6)

The magnitude of damage due to myocardial ischemia may be assayed based on the ST segment elevation. A relationship exists between the extent of ST segment displacement and the size of the infarction. The correlation between ST segment elevation and biochemical enzyme parameters are a matter of debate. There is no conclusive evidence available to prove the efficiency of ECG changes in the diagnosis of acute myocardial infarction. (7)

It was observed in experimental animals that CK clearance is not affected by blood pressure changes, heart rate and cardiac output. CK is not excreted in urine and the levels are not influenced by change in renal or hepatic blood flow. (8)

CKMB was detected initially at 6 hours or later after the onset of infarction. The mean time from the onset of symptoms to peak levels of CKMB was 32 hours, which is comparable to other reports. (8) Roberts also reported that CKMB disappears approximately 24 hours before CK levels return to normal. In

Table 1 : CK, CK MB, SGOT, α HBDH AND TROPONIN -T OF CONTROL GROUP

Subjects	Age (Years)	CK (U/L)	CKMB (U/L)	SGOT (U/L)	α HBDH (U/L)	Troponin-T (ng/ml)
Normal Blood Donors	37.6 $\pm$ 11.5	85.4 $\pm$ 30.5	8.5 $\pm$ 5.1	19.1 $\pm$ 9.4	39.2 $\pm$ 26.5	0.09 $\pm$ 0.18
Patients with polytraumatic injury	44.4 $\pm$ 15.2	102.6 $\pm$ 37.7	14.9 $\pm$ 8.6	25.6 $\pm$ 9.1	139.5 $\pm$ 43.7	0.09 $\pm$ 0.04
P Values for the above two group		< 0.001***	< 0.001***	< 0.001***	< 0.001***	0.5

Table 2 : CK, CK MB, SGOT, α HBDH AND TROPONIN –T IN PATIENTS WITH AMI

Time of the patients	CK (U/L)	CKMB (U/L)	SGOT (U/L)	α HBDH (U/L)	Troponin -T (mg/ml)
At the time of admission	218.4 $\pm$ 222.9	20.1 $\pm$ 5.1	42.4 $\pm$ 40.3	192.4 $\pm$ 141.7	2.04 $\pm$ 1.96
At 6 hours	826.5 $\pm$ 722.0	98.8 $\pm$ 72.7	60.6 $\pm$ 63.4	282.3 $\pm$ 232.2	4.97 $\pm$ 4.74
At 0 hr with 6 hours P values	< 0.001	< 0.001	< 0.001	< 0.050	< 0.001
At 14 hours	1668.1 $\pm$ 1563.2	187.2 $\pm$ 150.4	92.1 $\pm$ 91.2	365.2 $\pm$ 296.5	8.42 $\pm$ 7.94
At 0 hr with 14 hour P values	< 0.001	< 0.001	< 0.005	< 0.005	< 0.001
At 32 hours	2020.2 $\pm$ 1390.8	233.2 $\pm$ 181.2	129.1 $\pm$ 121.7	477.0 $\pm$ 385.1	11.56 $\pm$ 9.40
At 0 hr with 32 hours P values	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
At 48 hours	1373.0 $\pm$ 1550.4	163.1 $\pm$ 187.3	201.6 $\pm$ 172.4	515.2 $\pm$ 372.1	11.77 $\pm$ 7.72
At 0 hr with 48 hours P values	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
On 5 th Day	125.8 $\pm$ 34.6	15.2 $\pm$ 5.7	418.2 $\pm$ 280.8	741.0 $\pm$ 260.3	15.34 $\pm$ 7.83
At 0 hr with 5th day P values	< 0.005	< 0.100	< 0.001	< 0.001	< 0.001
On 7 th Day	80.2 $\pm$ 32.8	6.7 $\pm$ 4.6	557.3 $\pm$ 300.8	867.2 $\pm$ 194.1	17.58 $\pm$ 7.71
At 0 hr with 7th day P values	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001

the present study CKMB disappears approximately 48 hours after onset of symptoms. The average interval from onset of symptoms to appearance of peak values and disappearance of total CK, CKMB as well as the mean values for CK, CKMB were comparable to previous reports. (9)

Avery et al (1985) demonstrated that myoglobin a protein present in both skeletal and cardiac muscle is rapidly released into coronary vessels after successful reperfusion of injured myocardium. In all patients successfully undergoing coronary artery occlusion and reperfusion, blood CKMB increased rapidly after reopening of the vessel with systemic arterial peak occurring at 20 to 40 minutes. These early arterial peaks were followed by a rapid washout of myoglobin with half-time disappearance of 38 ± 3 minutes. (10)

The present study also revealed similar findings of CK and CKMB in patients who underwent successful coronary reperfusion for acute myocardial infarction produced by arterial occlusion of 99 to 100 %. In each case, plasma CK and CKMB rise rapidly after successful vessel reopening and peak level was followed by rapid plasma washout. The values for CK were more than 200 U/L and those of CK MB were more than 300 U/L. As the size of infarct shows an increase, the release of CK and CKMB also increase proportionately, thereby probably helping to get an idea of the magnitude of infarction. Gama (11) measured AST on three consecutive days after admission, in AMI patients and confirmed that the AST had a diagnostic sensitivity of 100% and specificity of 86 % although electrocardiograms of first two days of admission had a low diagnostic sensitivity (33.3%).

Yoneda et al found successful reperfusion by measuring AST after AMI. In our study also the washout of AST and α HBDH is more and remains elevated even on 7th day indicating reperfusion. (12)

Troponin T is a part of the regulatory system of the contractile complex of skeletal and heart muscle. It is expressed in two different isoforms namely skeletal muscle Troponin T and cardiac Troponin T. After the loss of integrity of the cell membrane, it is released into the circulation similar to Myoglobin or CK and CKMB. (13) Cardiac Troponin T is ordinarily undetectable in healthy individuals.

In the present study the serial changes of Troponin T values in AMI were matching with those of AST and α HBDH with the important exception that the Troponin T was elevated at the time of admission of the patient. (13) When the values for CK, CKMB and Troponin T were compared between normal blood donors and patients with polytraumatic injury, the difference was not significant (< 0.100, <0.005, > 0.500 respectively for the above three) Collinson et al had made the similar observations.(14)

The CTnT is unique and so can be differentiated from the isoforms in the skeletal muscle leading to a very high specificity for myocardial damage. (15) The wide time window from 4 hrs to 7 days or more than that and high sensitivity and specificity make CTnT suitable for detecting myocardial damage. Troponin T determination should prove to be an important tool for early risk assessment in-patients with unstable coronary artery disease. The maximum CTnT value

obtained during first 24 hr. provides independent and important prognostic information. (16)

The pattern of changes in the concentrations of AST and α HBDH showed variation from that of CK and CKMB. Both the AST and α HBDH were normal at the time of admission. The values for AST, α HBDH started rising at six hours and exhibited further rise at 14, 32 and 48 hours. When the estimations were done on 5th and 7th days their activities showed further increase. Troponin T in AMI was matching with those of AST and α HBDH with the important difference that the Troponin T was elevated at the time of admission of the patient whereas, the AST and α HBDH levels were well within the normal limits. The activity of cardiac Troponin T exhibited further rise at six, fourteen, thirty-two and forty eight hours. The values showed a further increase on fifth and seventh day.

Cardiac Troponin T has a wide time window from 4 hours to 7 days for detecting myocardial damage. The maximum cardiac Troponin T value obtained during the first 24 hours provides independent and important prognostic information. The increase of cardiac Troponin T was 20 times higher than the normal blood donors in AMI at the time of admission whereas that of CKMB was around two and half times in comparison to normal blood donors (17). Our values of CTnT are in agreement with a very recent study (18).

REFERENCES

1. Neumeir D. Activity kinetics and diagnostic significance in myocardial Infarction. Clinical chemica acta 1996; 73:445-51.
2. Hafkensheid JCM. J Clin chem clinical biochem 1979 ; 17 : 219.
3. Remppis A, Scheffaid T Karrer, O Zehelein, J Hamm, C Ketus, K. A. Assessment of reperfusion of the infarct zone after acute myocardial infarction by serial cardiac Troponin T measurement in serum. Br Heart J 1994 ; 71 : 3 : 242-6.
4. Lee T H, Rouan G W, Weiberg M. C. Sensitivity of routine clinical criteria for diagnosing myocardial infarction within 24 hrs of hospitalization. Ann Intern Med 1987; 106: 181-6.
5. Mc Queen MJ, Holder D, E L Moragi NRH. Assessment of accuracy of serial electrocardiograms in the diagnosis of myocardial infarction. Amm heat J 1983; 105: 258-6.
6. Rosano T G ,Claysan K J, Strandjord PE. Clinical Chem 1976; 22: 1078.
7. Krucaff N W, Green C E , Satler L F. Noninvasive detection of coronary Artery potency using continuous ST segment monitoring. Am J Cardiol 1986; 57: 916-2.
8. Roberts R Ishikawa Y. Enzymatic estimation of infarct size during reperfusion. Circulation 1983 ; 68 : Suppl-1: 83-9.
9. Galen R S. The enzyme diagnosis of myocardial infarction. Hum Pathol 1975 ; 56: 141-55.
10. Lott J A. Serum enzyme determination in the diagnosis of acute myocardial infarction. Hum Pathol 1984; 15: 706 -16.
11. Gama R, Swain D G. The effective use of cardiac enzyme and electrocardiogram in the diagnosis of acute myocardial infarction. Post grad Med J1990 ; 775 : 375-7.
12. Yondeda K, Katayarna Y, Takagi Y, Goni K. Early diagnosis and detection of successful reperfusion by mitochondrial AST/ soluable AST ratio after myocardial infarction. Rinsho-Byoti 1995 ; Jan 43-1 : 74-80.
13. Adams J E, Abendochain D R , Jaffe A.S. Biochemical markers of myocardial injury is MB creatinine kinase the choice for the 1996's ? Circulation 1993; 88: 750-53.
14. Colliension PO ,Chandar M A. Measurement of serum TnT , CKMB and total CK following arduous physical training. Ann. Clin. Biochem 1995 ; Sept. 2-32: 450-3
15. Ravkide J, Horder M, Gerhardt W, Liungdohi L et al. Diagnostic performance and prognostic value of serum Troponin T in suspected acute myocardial infarction. Scand J . Clin lab. Invest. 1993 ; 53 : 677-85.
16. Lindohi Bertil , Pervenge M B , Lass ,Wallentin. MD, Ph.D. For the FRISC study group relation between Troponin T and the rise of subsequent cardiac events in unstable coronary artery disease. Circulation: 1996 ; 93 :1651-7.
17. Shah Hitesh, Haridas N. Evaluation of clinical Utility of Serum Enzymes and Troponin –T in the Early Stages of Acute Myocardial Infarction. Indian Journal of Clinical Biochemistry.2003; 18 (2) : 93-101.
18. Reddy G. Chinnapu, Kusumanjali G, Sharada AHR, Rao Pragna. Cardiac Troponin – T and CKMB (Mass) levels in cardiac and non cardiac disease: Indian Journal of Clinical Biochemistry .2004; 19 (2) : 91-4.