Abstract

**Objective** Although heart-type fatty acid-binding protein (H-FABP) is a cardiac marker useful for early diagnosis of acute myocardial infarction (AMI), few data are available on its prognostic value. The objective of this study is to clarify the prognostic value of H-FABP in patients with a serious condition.

**Methods and Patients** We conducted a prospective study of 617 patients who presented to the emergency department with a serious condition. The H-FABP levels on arrival at the emergency department were divided into four groups using their quartiles. The endpoint was death from any causes in-hospital.

**Results** H-FABP ranged from 1.2 to 2,300 ng/ml, with a median of 19.9 ng/ml, a 25%-value of 6.7 ng/ml and 75%-value of 54.0 ng/ml. The unadjusted rate of the mortality increased progressively with increasing H-FABP quartile point (11% for quartile-I, 22% for quartile-II, 36% for quartile-III, and 38% for quartile-IV; p<0.001). After adjustment for age, gender, systolic blood pressure and the presence or absence of cardiovascular disease, H-FABP was the independent factor to predict the mortality.

**Conclusion** H-FABP has proven to be an independent factor for prognosis in patients with a serious condition on arrival at the emergency department.

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**Key words:** heart-type fatty acid-binding protein (H-FABP), false-positive test, acute myocardial infarction (AMI), prognosis

Introduction

Human heart-type cytoplasmic fatty acid-binding protein (H-FABP) is a low molecular weight protein (14.9 kDa) that takes part in fatty acid transport and buffering, and is distributed predominantly in the cytosol of myocardium. H-FABP occurs in part in skeletal muscles as well, and its content in the myocardium is reportedly 3 to 10 times as high as in the skeletal muscle (1, 2). H-FABP became recognized as a cardiac marker in the 1990s (3–6). Troponin and CK-MB isozyme have been reported to be of low diagnostic sensitivity for early after onset of acute myocardial infarction (AMI), their approximate diagnostic sensitivity being 20% at 2 hours, 40% at 4 hours, and 60% at 6 hours after the onset (7, 8). As H-FABP appears in the circulating blood as early as 1.5 hours after onset of AMI, it is feasible to make an early diagnosis of AMI within 3 hours after onset (3, 9). H-FABP liberated into the bloodstream is largely excreted from the kidneys and is eliminated from the blood in 24–36 hours if there is no renal dysfunction (10, 11). Its dynamic features of liberation into circulating blood resemble those of myoglobin but are characterized by high specificity to myocardium. Nevertheless, H-FABP is slightly inferior to troponin and CK-MB in diagnostic specificity for AMI and has been described to be increased in other cardiogenic disorders, renal dysfunction, and disorders associated with skeletal muscle damage (1, 10–13).

A simplified rapid assay kit (Rapicheck® H-FABP, Dainippon Pharmaceutical Company, Osaka) that enables testing as fast as within 15 minutes has been developed (14) and has become available at sites of emergency department since 2002 and the clinical significance has been proven (15). However, reports of studies assessing disorders showing false-positive tests for H-FABP or assessing whether H-FABP has any applicability for prognosis are few. This
prospective study represented an attempt to clarify the prognostic value of H-FABP in the management of patients with a serious condition under emergency medical care.

Methods

Study Population

The subjects of this study were emergency patients with a serious condition transported by ambulance to the emergency department. A definite diagnosis was made through integration of clinical features, laboratory findings and various imaging techniques, the diagnosis of AMI and that of unstable angina (UA) being based on the redefined ESC/ACC criteria, revised in September 2000 (16), rather than the conventional WHO criteria (17). Infants, children, and patients whose venous blood samples could not be withdrawn prior to emergency medication, and patients (or their relatives) from whom consent to participate in the study could not be obtained were excluded from the study.

Measurement of H-FABP Levels

Blood samples for H-FABP were collected from the peripheral vein on arrival and before emergency medication, and assayed by the two-step sandwich ELISA using two anti-human H-FABP-specific monoclonal antibodies. The assay system is based on the following principle. H-FABP in a serum sample is trapped with anti-human H-FABP-specific monoclonal antibody of mouse origin immobilized onto wells of the assay plate. Then POD-labeled anti-human H-FABP-specific monoclonal antibody of mouse origin is added. Adding a substrate to the resultant immune complex comprised of these three reagents and measuring the absorbance at 492 nm (18).

Study Endpoint and Statistical Analysis

The study endpoint was death from any causes in-hospital within 30 days. Assay values of H-FABP were divided into four groups using quartiles of the median, 25% value and 75% value, to analyze the data for the proportion of each disorder and the study endpoint.

All continuous variables were expressed as the mean±SD. Continuous variables of patient background characteristics were tested using one-factor ANOVA, and categories were assessed with the chi-square test for independence. The chi-square test for independence was employed for statistical assessment of the distribution of each disorder and the outcome as to inter-group differences among H-FABP quartiles. Logistic regression analysis was adopted in determining whether H-FABP represented an independent factor or not. Also, multiple regression analysis was adopted in determining the correlation between H-FABP and other severity indexes. The cut-off point for the prognostic differentiation of cardiovascular diseases was tested using a receiver-operating characteristic (ROC) curve. Any differences found were considered statistically significant if p<0.05.

Results

Baseline Characteristics

Of 885 patients transported by ambulance as emergency patients with serious condition to the emergency department during the 15-month period from October 2000 to December 2001, 617 patients (69.7%) fulfilling the enrollment criteria (464 men; mean age, 57±19 years) were admitted to the study. As seen in Table 1, cardiovascular disease accounted for 32% of all cases (AMI, 13% of all cases; heart failure, 7%; UA, 6%; and aortic disease, 3%), cardiopulmonary arrest (CPA) on arrival for 19% (cardiogenic, 15%; and non-cardiogenic, 4%), trauma for 17%, cerebrovascular disease for 9%, and others for 22% (e.g. gastrointestinal and hepatobiliary disease such as gastrointestinal hemorrhage, fulminant hepatitis and severe acute pancreatitis, 49 cases; drug poisoning, 26; renal failure, 17; epileptic seizures, 14; severe respiratory failure such as bronchial asthma and acute lung injury, 9; sepsis, 8; hypoglycemia, 6; and burns, 2).

The distribution of serum H-FABP levels showed a range from 1.2 to 2,300 ng/ml, with a median of 19.9 ng/ml, a 25%-value of 6.7 ng/ml, and a 75%-value of 54.0 ng/ml (mean, 71±189 ng/ml). Serum H-FABP values of 1.2 to <6.7 ng/ml were taken as the first quartile (Q-I), 6.7 to <19.9 ng/ml as the second quartile (Q-II), 19.9 to <54.0 ng/ml as the third quartile (Q-III), and 54.0 to <2,300 ng/ml as the fourth quartile (Q-IV).

Baseline characteristics of the patients in these four groups according to H-FABP quartiles are summarized in Table 2. The CPA cases were excluded from analysis of systolic blood pressure, heart rate and respiratory rate on arrival. There were significant inter-group differences with respect to age, systolic blood pressure, pH and base excess (BE).
**H-FABP as a Marker of Prognosis**

**Table 2. Characteristics According to the Quartile of H-FABP Level**

<table>
<thead>
<tr>
<th></th>
<th>Q-I</th>
<th>Q-II</th>
<th>Q-III</th>
<th>Q-IV</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of patients</td>
<td>155</td>
<td>154</td>
<td>155</td>
<td>153</td>
<td></td>
</tr>
<tr>
<td>Age, yr</td>
<td>52±17*</td>
<td>58±18</td>
<td>61±18</td>
<td>58±19</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Male sex, no. (%)</td>
<td>111 (72)</td>
<td>118 (77)</td>
<td>119 (77)</td>
<td>116 (76)</td>
<td>0.3</td>
</tr>
<tr>
<td>SBP, mmHg†</td>
<td>138±37</td>
<td>144±49</td>
<td>127±44</td>
<td>123±38</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>HR, bpm†</td>
<td>86±26</td>
<td>97±24</td>
<td>94±27</td>
<td>94±30</td>
<td>0.06</td>
</tr>
<tr>
<td>RR, bpm†</td>
<td>22±12</td>
<td>22±6</td>
<td>21±6</td>
<td>23±9</td>
<td>0.6</td>
</tr>
<tr>
<td>pH</td>
<td>7.39±0.13</td>
<td>7.32±0.45</td>
<td>7.26±0.21</td>
<td>7.30±0.20</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>BE</td>
<td>−0.2±4.8</td>
<td>−2.5±6.1</td>
<td>−5.8±8.2</td>
<td>−5.3±8.0</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

SBP: systolic blood pressure, HR: heart rate, RR: respiratory rate, BE: base excess. †Plus-minus values are means±SD. *Plus-minus values are means±SD. †Patients with cardio-pulmonary arrest were excluded.

Patients with higher H-FABP groups (Q-III, Q-IV) were older ages and had a lower systolic blood pressure, pH and BE, compared to patients in lower H-FABP groups (Q-I, Q-II).

**Distribution according to H-FABP Quartiles**

Patients with serum H-FABP levels deviating from the conventional AMI diagnostic criterion (6.2 ng/ml) accounted for 76% of all cases. Figure 1A shows the number of patients with each cardiovascular disease divided into H-FABP quartiles. The number of patients with AMI increased stepwise with increasing quartile points (p<0.005) and 89% of patients with AMI had deviations from the conventional reference value of 6.2 ng/ml. The number of patients with heart failure (HF) also exhibited significant differences among the quartiles, with a peaked pattern of distribution (p<0.005), and 91% had deviations from the value of 6.2 ng/ml. In trauma, most of patients also had elevated H-FABP levels but there was no significant inter-quartile difference, but 69% had deviations from the value of 6.2 ng/ml.

Figure 1B shows the number of patients with CPA and non-cardiovascular disease divided into H-FABP quartiles. The number of CPA cases increased stepwise with increasing quartile points (p<0.001), and 91% had deviations from the value of 6.2 ng/ml. In trauma, most of patients also had elevated H-FABP levels but there was no significant inter-quartile difference, and 86% had deviations from the value of 6.2 ng/ml. On the other hand, the number of patients with cerebrovascular disease decreased stepwise with increasing quartile points (p<0.005), and 68% had deviations from the value of 6.2 ng/ml. The patients with renal failure were concentrated in Q-III and Q-IV (p<0.001), and 100% had deviations from the value of 6.2 ng/ml.

**Outcome**

A total of 166 patients (27%) died in-hospital, and the H-FABP levels were higher among such patients than among those who survived until hospital discharge (mean±SD, 111 ±244 ng/ml vs. 56±162 ng/ml, p<0.001). In Fig. 2, the study endpoint, in-hospital mortalities for the four H-FABP groups of the overall patient population (left) and those excluding CPA cases (right) are shown. The former significantly increased stepwise with increasing quartile points, i.e., 11% for Q-I, 22% for Q-II, 36% for Q-III, and 38% for Q-IV (p<0.001). In the population excluding CPA cases, inhospital mortalities for Q-II, Q-III, Q-IV were approximately twice as high as Q-I, although there was no significant difference among the quartiles.

In the multiple logistic regression analysis for independent predictors of the study endpoint including age, gender, systolic blood pressure and the presence or absence of cardiovascular disease, the increasing quartiles of the H-FABP levels remained significantly associated with increased risk of death. Figure 3 shows that the adjusted odds ratios for the study endpoint in the second, third, fourth quartiles of H-FABP were 2.47 (95% CI, 1.29 to 4.74, p<0.01), 4.85 (95% CI, 2.60 to 9.09, p<0.001) and 5.49 (95% CI, 2.96 to 10.2, p<0.001), respectively.

In addition, multiple regression analysis was performed to evaluate the correlation between H-FABP and other severity indexes including systolic blood pressure, heart rate, BE and blood glucose. As seen in Table 3, H-FABP was significantly correlated with systolic blood pressure (p=0.003) and BE (p=0.04). Because infarct size, as previously known, is correlated with prognosis in patients with AMI, it is essential to evaluate the relationship between H-FABP and prognosis in patients without AMI. Here, the in-hospital mortalities for the four H-FABP groups of patients excluding AMI from overall patient population (n=534) significantly increased stepwise with increasing quartile points, i.e., 11% (16/143) for Q-I, 23% (32/141) for Q-II, 42% (55/130) for Q-III, and 48% (57/120) for Q-IV (p<0.001). H-FABP was significantly correlated with systolic blood pressure (p=0.006) and BE (p=0.007) even in this population.

In sub-analysis, in-hospital mortalities for the H-FABP quartile groups of patients focused on fatal cardiovascular diseases producing acute chest pain [confined here to 141 cases of acute coronary syndrome (ACS), acute aortic
dissection (AAD), and acute pulmonary thromboembolism (PE) were shown in Fig. 4. There was a stepwise increase in mortality with increasing H-FABP quartiles (p<0.05). To clarify the prognostic accuracy of H-FABP, the receiver-operating characteristics (ROC) curve for death was evaluated.

Figure 1. A) H-FABP distribution in cardiovascular diseases. The H-FABP level increased progressively with increasing quartile points in patients with AMI (p<0.005). In patients with heart failure (HF), there were significant inter-quartile differences, with a peaked pattern of distribution (p<0.005). In patients with unstable angina (UA), the H-FABP level decreased progressively with increasing quartile points (p<0.001); while no significant inter-quartile difference was observed in patients with aortic disease (AD). B) H-FABP distribution in CPA and non-cardiovascular diseases. The H-FABP level showed an escalation with increasing quartile points in patients with CPA (p<0.001). The majority of patients of trauma also displayed elevations but no significant inter-quartile difference. In patients with cerebrovascular diseases, on the other hand, there was a stepwise decrease with increasing quartile points (p<0.005). In patients with renal failure were concentrated in Q-III and Q-IV (p<0.001), no significant inter-quartile difference was observed in patients with sepsis.

Table 3. Correlation between H-FABP and Variable Severity Indexes in Multiple Regression Analysis

<table>
<thead>
<tr>
<th>Variables</th>
<th>R</th>
<th>95% CI</th>
<th>P-value</th>
</tr>
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<tbody>
<tr>
<td>Systolic blood pressure</td>
<td>-0.407</td>
<td>-0.675 to -0.139</td>
<td>0.003</td>
</tr>
<tr>
<td>Heart rate</td>
<td>0.238</td>
<td>-0.160 to 0.636</td>
<td>0.240</td>
</tr>
<tr>
<td>Blood glucose</td>
<td>0.091</td>
<td>-0.056 to 0.238</td>
<td>0.226</td>
</tr>
<tr>
<td>Base excess</td>
<td>-2.288</td>
<td>-4.467 to -0.109</td>
<td>0.040</td>
</tr>
</tbody>
</table>

(PE) were shown in Fig. 4. There was a stepwise increase in mortality with increasing H-FABP quartiles (p<0.05). To clarify the prognostic accuracy of H-FABP, the receiver-operating characteristics (ROC) curve for death was evaluated.
determined by analysis of data from these 141 patients. The area under the ROC curve (AUC) was 0.76 (95% CI, 0.65 to 0.87) (Fig. 5). When the cut-off point of H-FABP was 47.5 ng/ml, the sensitivity (64%) + specificity (74%) of the test was maximal, and the accuracy was 74% for the identification of death.

Discussion

The present study demonstrated that 76% (469/617) of the study patients in a serious condition upon arrival at the emergency department had deviations from the conventional AMI diagnostic value of 6.2 ng/ml and the risk of death showed an escalation with increasing serum H-FABP levels. This trend was evident not only in the overall patient population but in patients with cardiovascular diseases as well. A multiple logistic regression analysis showed H-FABP to be an independent factor for death in-hospital. We determined the optimal cut-off point of H-FABP regarding death in-hospital to be 47.5 ng/ml in patients with cardiovascular disease.

H-FABP is recognized as a useful cardiac marker for making the diagnosis of hyper acute phase of AMI. But the survival prognostic value is still unknown. The first report of the relationship between H-FABP and prognosis was published in 2003 (19). It showed H-FABP can predict the early coronary intervention within the first 7 days in patients with suspected ACS. It did not mention the survival prognosis, so this study is the first report on survival prognosis using H-FABP. Because a good correlation of the myocardial infarct size with H-FABP level has been documented in previous studies (4, 5), it is reasonable that H-FABP is correlated with the prognosis in patients with AMI. But in our study, H-FABP was also correlated with the prognosis in patients excluding AMI (n=534). As H-FABP elevation is caused by damage to the myocardium, skeletal muscle or both, H-FABP might reflect invasiveness of the myocardium or skeletal muscle. In addition, H-FABP was found to be significantly correlated with systolic blood pressure and BE in patients excluding AMI. These parameters, which are indexes of severity, suggest the degree of invasiveness. Once critical situations such as shock, multiple organ dysfunction syndrome (MODS), and systemic inflammatory response syndrome (SIRS) occur, skeletal muscle and the gastrointestinal system fall into hypoperfusion state earlier than other important organs. It is thought that ischemia is caused by microvascular dysfunction (20, 21). Therefore, the higher the H-FABP level, the greater the degree of invasiveness of myocardial and/or skeletal muscle damage and the poorer the prognosis.

There were 9 patients with AMI whose H-FABP levels were less than the reference value of 6.2 ng/ml. They were false-negative cases for AMI. Two of them were so-called microinfarction cases in which serial determinations of cardiac markers over time revealed deviations of troponin and H-FABP levels from the reference value. In the remaining 7 patients, they had overt chest pain and ECG abnormalities but blood samples were taken within 1 hour post-onset, therefore H-FABP had not become elevated yet. The
deviations from the reference value were confirmed by retests of H-FABP 3 hours after onset. These findings suggest that serial determinations of H-FABP over time are required in cases suspected AMI with normal H-FABP within 1 hour post-onset.

H-FABP elevations were also frequently noted among patients with other cardiovascular diseases. High H-FABP levels were observed in a considerable proportion of patients with heart failure, consistent with several reports documenting H-FABP elevations in heart failure patients (12, 13). These reports stated that H-FABP is useful for assessing the severity of the prognosis in patients with heart failure. Conversely, the number of patients with UA decreased stepwise with increasing quartile points of H-FABP. It has been previously reported that elevated H-FABP is found in some of the patients with UA (3, 6). The cases of microinfarction were dealt as AMI in this study according to ESC/ACC re-definition (16), but H-FABP was found to be deviated from the reference value (6.2 ng/ml) in 30% (11 cases) of all UA cases. This may reflect the presence of more minute myocardial damage, as H-FABP has been reported to have greater diagnostic sensitivity than troponin (22, 23). Of note, there were 3 renal dysfunction cases (serum creatinine >1.5 mg/dl) in 11 UA cases showing H-FABP elevation. In the patients with aortic disease, H-FABP elevation was considered to be due not only to myocardial damage caused by aortic valve regurgitation or cardiac tamponade, but also to organ ischemia or skeletal muscle damage caused by vascular lesions.

Much remains to be clarified as to mechanisms in CPA cases, and it is still unclear whether H-FABP values are elevated or not. We did not daringly exclude the CPA cases in this study, and also examined the details of H-FABP values for the CPA cases. H-FABP elevation was more frequent among CPA cases than in AMI, with a stepwise increase which resembled AMI in distribution. According to an analysis of the data from CPA cases by the Utstein style (24), the causes of CPA were cardiogenic in more than 60%, and additionally with AMI accounting for a noticeable percentage of these cardiogenic CPA (25–28). Therefore, more advanced myocardial damage leads to a fatal situation. In patients with aortic disease, H-FABP elevation was considered to be due not only to myocardial damage caused by aortic valve regurgitation or cardiac tamponade, but also to organ ischemia or skeletal muscle damage caused by vascular lesions.

Study Limitations

In the present study, as we considered a simple study design, the H-FABP levels were measured only one time on arrival, however peak H-FABP levels by serial determinations over time would provide further useful information. This was not a multicenter study for the measurement of serum H-FABP and the analysis of the data, and the diagnosis of each patient in the emergency department depended on several clinical physicians’ experience. Furthermore, emergency medical service personnel made the judgment of the serious condition instead of clinical specialists of emergency medicine. These factors might cause a slightly produce bias in a study results.

Conclusions

In patients with serious condition on arrival at the emergency department, the level of H-FABP can provide valuable information regarding the risk stratification of death.

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References


