Relationship between heart-type fatty acid-binding protein levels and coronary artery disease in exercise stress testing: an observational study

Egzersiz stres testinde kalp-tipi yağ asidi-bağlayıcı protein düzeyleri ile koroner arter hastalığı arasındaki ilişki: Gözlemsel bir çalışma

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ABSTRACT

Objective: Although, there has been great improvement on the diagnosis and early treatment of acute coronary syndromes, especially in terms of myocardial damage biochemical markers, we do not have a specific marker yet, for using the diagnosis of stable coronary artery disease (CAD). This study aimed to evaluate the relationship between CAD and the changes of heart-type fatty acid binding protein (H-FABP) levels before and after exercise stress testing (EST).

Methods: A total of 47 patients were enrolled in this observational study. Of 47 patients, 21 had normal coronary anatomy; the remaining 26 patients had coronary lesions over 70% in at least one major coronary artery. All patients performed EST. Along with this, H-FABP levels before EST and at peak exercise, 1st hour, 3rd hour (3h), were measured in all patients. Differences among the measurements were evaluated through the Friedman test and Wilcoxon test, and the Bonferroni correction was applied to determine which measurement caused the difference.

Results: Contrary to expectations, the means of the H-FABP values measured at particular intervals for each group tended to decline from the basal level to the 3h level. When the difference between the 3h measurement and the basal level was compared in each group, the decreasing was statistically significant in both groups ($p<0.05$). A statistically significant decrease at the 3h measurement compared to the basal level in the CAD group was more apparent than in the control group (2.790±2.569ng/ml vs. 0.837±2.070ng/ml, $p=0.009$).

Conclusion: We found that H-FABP levels did not increase during EST and contrary to expectation, were inclined to decrease. We thought that decreasing H-FABP levels likely resulted from exercise-induced proteinuria. *(Anadolu Kardiyol Derg 2011; 11: 685-91)*

Key words: Coronary artery disease, heart-type fatty acid binding protein, exercise stress testing

ÖZET

Amaç: Akut koroner sendrom tanısı ve erken tedavisinde özellikle miyokardiyal hasar belirteçlerinde sağlanan ilerleme ile önemli bir mesafe alınmasına rağmen stabil koroner arter hastalığı (KAH) tanısında kullanılabilecek özel bir markır henüz yoktur. Bu çalışmada egzersiz stres testi (EST) öncesi ve sonrası H-FABP düzeylerindeki değişim ile koroner arter hastalığı arasındaki ilişki araştırılmıştır.

Yöntemler: Gözlemsel çalışmamıza en az bir majör koroner arterinde %70 ve üzerinde darlığı olan 26 hasta ve koroner arterleri normal bulunan 21 hasta olmak üzere toplam 47 hasta alındı. Tüm hastalara EST yapıldı. Tüm hastalarda bazal, pik egzersiz, 1. saat ve 3. saatte H-FABP ölçümü yapıldı. Ölçümler arası farka Friedman Testi ile bakıldı, farkın hangi ölçümden kaynaklandığını bulmak için Bonferroni düzeltmeli Wilcoxon Testi kullanıldı.

Bulgular: Her bir grupta belirlenen zaman aralığında ölçülen H-FABP değerleri ortalamaları beklenenin aksine bazaldan üçüncü saate azalma eğiliminde idi. Üçüncü saat ile bazal ölçümler arasındaki farklar (fark 3) grup içinde karşılaştırıldığında her iki grupta da azalma anlamlı idi ($p<0.05$). Her iki grubun 3. saat ile bazal arasındaki farkları (fark 3) karşılaştırıldığında da KAH grubunda 3. saat ile bazal ölçüm arasındaki fark kontrol grubundan daha fazlaydı (2.790±2.569ng/ml, 0.837±2.070ng/ml, $p=0.009$).

Sonuç: Çalışmamızda H-FABP düzeylerinin EST'de yükselmediğini, beklenenin aksine bazal değerlere göre azaldığını tespit ettik. H-FABP düzeylerindeki azalmanın egzersize bağlı proteinüri nedeniyle oluştuğunu düşünmekteyiz. *(Anadolu Kardiyol Derg 2011; 11: 685-91)*

Anahtar kelimeler: Koroner arter hastalığı, kalp tipi yağ asidi bağlayıcı protein, egzersiz stres testi

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Introduction

There has been a great improvement on the diagnosis of acute coronary syndrome (ACS) in terms of myocardial damage biochemical markers; however, there are still some deficiencies, especially in the evaluation of patients with asymptomatic coronary artery disease (CAD) or chest pain. Exercise stress testing (EST) is the most frequently used diagnostic method for diagnosing stable CAD. Unfortunately, EST has some deficiencies due to its relatively low specificity and sensitivity. A meta-analysis of 147 studies investigating the diagnostic accuracy of EST demonstrated that the mean sensitivity was 68% (23-100) and that the mean specificity was 77% (17-100). However, these figures varied widely among the studies (1).

Heart-type fatty acid binding protein (H-FABP) is a cytoplasmic protein having a relatively small molecular weight (15kDa) (2). It constitutes 2-5% of the total cytosolic protein in the heart muscle (3). It was shown to be a sensitive and early marker of myocardial damage in ACS in many studies (4-8). In addition, it was indicated that high plasma H-FABP levels in the different processes of myocardial damage, such as heart failure, have been associated with poor prognosis and cardiovascular events in the future (9-12). Although many studies have been conducted on H-FABP levels in ACS and other cardiac pathologic courses accompanying myocardial damage, studies evaluating whether these levels are affected by temporary myocardial ischemia in patients with stable CAD are very rare.

A study of 47 patients suspected of having CAD demonstrated that H-FABP levels measured before and 3 and 5 hours after bicycle exercise stress testing were not elevated and did not differ between groups having CAD or not, as determined by angiography (13). It was reported that H-FABP levels started increasing 1 hour following myocardial cell damage, peaked between 6th and 8th hours, and then returned to normal between 24th and 30th hours (7, 14). When it is thought the fact that temporary myocardial damage occurring during EST appears less than in ACS and H-FABP is removed from plasma quickly by the kidneys, EST may be more effective for detecting increases of this marker, assuming that measurements (to compare to this study) can be performed in early hours.

In this study, designed to confirm the above hypothesis, we aimed to evaluate whether myocardial ischemia induced by EST increases H-FABP levels. In addition, if myocardial ischemia induced by EST does increase H-FABP levels, we aimed to assess whether these findings, in combination with EST data, can improve the diagnostic performance of EST.

Methods

Study design and protocol

A total of 47 patients were enrolled in this prospective observational study. While 21 of those having normal coronary anatomy were accepted as the control group, the remaining 26 patients had coronary lesions over 70% in at least in one major coronary artery. All of the patients were selected consecutively from those who agreed to participate in the study and have undergone coronary angiography at the Department of Cardiology at the Selçuk University School of Medicine between 2008 December and 2009 November. Two patients with normal coronary anatomy were excluded from the study because blood samples could not be taken after EST. The patients having the following criteria were not included in this study owing to the fact that they might affect the potency of EST as well as the levels of the biochemical markers to be utilized.

Exclusion criteria:

- Having ACS in the last 6 weeks,
- Pericarditis and myocarditis,
- Serious left ventricle hypertrophy,
- Mild-to-severe mitral and aortic valve disease,
- Left ventricle systolic dysfunction (ejection fraction lower than 45%),
- Kidney and liver insufficiency,
- Severe lung disease and significant anemia,
- Malignity,
- Peripheral arterial disease,
- Known skeletal muscle disease or having engaged in heavy exercise in the last week,
- Any contraindication for performing EST.

The study has been approved by Ethics Committee of Selçuk University School of Medicine. All patients participating in this study provided informed consent.

Basal clinical examinations

All information about the patients, such as anamnesis, physical examination findings, history of previous anti-ischemic drug usage (beta blockers and long acting nitrates), demographic data (age, gender, weight, height, and risk factors for atherosclerosis), and various laboratory parameters [fasting glucose, urea, creatinine, sodium, potassium, aspartate aminotransferase (AST), alanine aminotransferase (ALT), hemoglobin, white blood cell count, fasting lipid profile] were registered.

Exercise testing and study protocol

Treadmill EST with an X-SCRIBE (Mortara INSTRUMENT, Milwaukee, WI, USA) device was applied to patients in accordance with Bruce's protocol.

A 4-ml blood sample for the measurement of cardiac troponin I (cTnI) and H-FABP was taken just prior to EST (basal) and then peak exercise (10 min after termination of EST), 1st hour, and 3rd hour after the termination of EST. A vascular access catheter located in an antecubital vein was used for obtaining blood samples. Blood samples were kept for 30 min to allow the blood to clot and then centrifuged for 12 min at 3000 revolutions per minute. The separated serum samples were put into Eppendorf tubes and stored at -80°C until processed biochemically.

Cardiac TnI levels before EST and at 3rd hour, along with H-FABP levels before EST, peak exercise (10 min after termination of EST) and at 1st hour and 3rd hour, were measured from the samples. EST recordings were evaluated by a cardiologist (at the end of the study) who was unaware of the results of the coronary angiography. All data obtained were compared between the groups and afterward compared within each group before and after the exercise for different time periods. The relation of changes in the parameters with EST findings was analyzed.

Biochemical analysis of cTnI and H-FABP

Cardiac TnI measurement was made using the ELISA technique from the basal (just before EST) and 3rd hour samples (cTnI ELISA kits - DRG International Inc, USA, Cat No: EIA 2952). The reference value for healthy individuals given by the technique is <0.5 ng/ml; however, the clinical cutoff value for acute myocardial infarction is 1.5 ng/ml Inter- and intra-study coefficient of variation (CV) values were 4% and 4.6%, respectively.

H-FABP levels were also measured using the ELISA technique from the basal, peak exercise (10 min after termination of EST), 1st hour, and 3rd hour samples. H-FABP ELISA (BIOCHECK Inc, USA, Cat No: BC-1123) kits were used to analyze the H-FABP levels. The minimum concentration determined by this technique is 0.25 ng/ml, the reference value for healthy individuals is 1.6-19 ng/ml, and the clinical cutoff value for acute myocardial infarction is 19 ng/ml. The inter- and intra-study CV values were 8% and 10.9%, respectively.

Statistical analysis

Statistical analysis of the study was performed with SPSS Version 13.0 (SPSS Inc., Chicago, Illinois, USA). The descriptive results are presented with mean±standard deviation and percentages. They were appropriate for normal distribution analyses. The comparison between the groups was performed with an unpaired Student's t-test. The Mann-Whitney U test was used to analyze the continuous variables not meeting normal distribution. Differences among the repeated measurements were evaluated through the Friedman test and Wilcoxon test, and the Bonferroni correction was applied to determine which measurement caused the difference. A Chi-square test was used to analyze categorical data. P values <0.05 were accepted as statistically significant.

Results

Basal characteristics (Table 1)

The number of male patients in the CAD group was higher. There was no difference between the CAD group and the control group (patients having normal coronary anatomy) in terms of age, body mass index, hypertension, family history and hyperlipidemia. The incidence of diabetes mellitus (DM) and smoking in the CAD group was significantly higher. The rate of having typical angina pectoris in the patients' backgrounds in the CAD group was obviously higher. Laboratory measurements of creatinine and hemoglobin were also similar for both groups. While there was no difference found between the two groups in terms of having a history of hyperlipidemia, low-density lipoprotein values were lower in the CAD group. On the other hand, the statin usage rate was higher in the CAD group.

EST results (Table 2)

No complications occurred during EST. The maximum heart rate reached during EST was lower in the CAD group than in the control group (128 and 143, respectively). There was no difference between basal and maximum exercise systolic blood pressure values, and the maximal MET values achieved were the same for both groups. Fifteen patients in the CAD group had a history of angina pectoris; however, no patient in the control group had a history of angina. When the electrocardiographic findings of the patients were evaluated, no ST segment elevations occurred in either group. On the other hand, ST segment depression ≥1mm occurred in 19 patients in the CAD group and in 10 patients in the control group, but this difference was not statistically significant. The characteristic of ST depression was more prominently downsloping in the CAD group. However, the number of upsloping ST depression was considerably higher in the control group. The comparison of EST results is summarized in Table 2.

H-FABP levels before and after EST (Tables 3, 4)

There was no difference in the basal cTnI measurements between the two groups (0.494ng/ml ±0.19 and 0.465ng/ml±0.19

Table 1. The demographic and clinic characteristics of the patients

Variables	Control group (n=19)	CAD group (n=26)	p*
Age, years	56.1±10.95	58.8±7.83	0.33
Male gender, n (%)	9 (31)	20 (69)	0.04
Body mass index, kg/m^2	29.1±3.59	27.80±4.13	0.30
Hypertension, n (%)	9 (47.4)	16 (61.5)	0.345
Diabetes mellitus, n (%)	2 (10.5)	11 (42.3)	0.02
Smoking, n (%)	6 (31.6)	16 (61.5)	0.04
Family history, n (%)	6 (31.6)	9 (34.6)	0.830
History of hyperlipidemia, n (%)	9 (47.4)	16 (61.5)	0.345
History of typical angina, n (%)	11 (57.9)	23 (88.5)	0.018
Q wave in basal ECG, n (%)	0 (0)	10 (38.5)	0.002
Creatinine, mg/dL	0.89±0.25	0.93±0.19	0.558
Low-density lipoproteins, mg/dl	132.00±40.48	108.00±34.00	0.040
Hemoglobin, gr/dL	13.20±1.44	13.72±1.32	0.276
Beta blocker usage, n (%)	6 (31.6)	16 (61.5)	0.047
Long-acting nitrate usage, n (%)	1 (5.3)	3 (11.5)	0.465
Statin usage, n (%)	4 (21.1)	17 (65.4)	0.003

Data are presented as number (percentage) and mean±SD values
*Chi-square test and unpaired Student-t test
CAD - coronary artery disease

p=0.827, respectively). However, the cTnI values 3 h after EST were substantially higher in the CAD group (0.854ng/ml±0.83 and 0.504ng/ml±0.24 p=0.027, respectively). This increase in cTnI level never reached the cutoff value needed for myocardial infarction.

As for H-FABP measurements, there were no differences between the CAD and control group for all of basal values (before EST), peak exercise values (10 min into exercising), and values at 1 hour and 3 hour after EST. None of these measurements reached the cutoff H-FABP value accepted for diagnosis of myocardial infarction.

The means of the H-FABP values measured at particular intervals for each group tended to decline from the basal level to the 3rd hour level (Fig. 1). When comparing, peak exercise measurement to the basal level (difference 1), the 1st hour measurement to the basal level (difference 2), and the 3rd hour measurement to the basal level (difference 3), differences 2 and 3 in the CAD group were statistically significant (p=0.003 and p<0.0001, respectively). However, only difference 3 in the control group was statistically significant (p=0.024).

When differences 1, 2, and 3 were compared between the two groups, difference 3 in the CAD group was higher than in the control group (2.790±2.569ng/ml, 0.837±2.070ng/ml, p=0.009). In other words, a statistically significant decrease at the 3rd hour measurement compared to the basal level in the CAD group was more apparent than in the control group. Comparisons of the differences between H-PABP measurements for both groups are shown in Table 4 and Figure 2.

No differences were observed at basal and peak levels and at the 1st hour and 3rd hour measurements during EST for each group when comparing the H-FABP levels between the patient groups that had ST depression and those that did not.

Discussion

This study shows that H-FABP levels did not increase due to myocardial ischemia induced by EST and contrary to expectation, were inclined to decrease in both CAD and control group after EST. Possible mechanism and importance of H-FABP levels decreasing have discussed at the following.

Although EST is the most frequently used diagnostic method for diagnosing stable CAD, it has some deficiencies because of its relatively low sensitivity and specificity. Different conclusions have been drawn from the literature depending on the hypothesis that myocardial ischemia induced by EST may affect the plasma level of biochemical markers and this condition may improve the diagnostic accuracy of EST. Myocardial ischemia and injury markers, such as cTn (13, 15, 16), glycogen phosphorylase BB (13), and IMA (17, 18), were applied in these studies and controversial results were obtained in general. Additionally, a relationship was established between the existence of CAD and the measurement of biochemical markers reflecting myocardial stress, such as BNP and NT-proBNP, at various time intervals before and after EST (19, 20).

Table 2. The comparison of EST results

Variables	Control group (n=19)	CAD group (n=26)	p*
Maximum heart rate, beats/min	143±20.95	128±18.82	0.011
Maximal MET, unit	9.7±2.10	9.3±2.21	0.555
ST segment depression ≥1 mm, n (%)	10 (52.6)	19 (73.1)	0.157
ST segment depression, n (%)			
Downsloping	0 (0)	13 (68.4)	<0.0001
Upsloping	5 (50)	0 (0)	<0.0001

Data are presented as number (percentage) and mean±SD values
*Chi-square test and unpaired Student-t test
CAD - coronary artery disease, EST - exercise testing, MET - metabolic equivalent

Table 3. Comparisons of serial H-FABP measurements during and after EST

HAFBP, ng/ml	Control group (n=19)	CAD group (n=26)	p#
Basal	6.77±2.29 6.3 (10.7,3.4)	7.65±3.15 7.02 (17.9,3.4)	0.402
Peak	6.72±2.01 6.5 (9.7,3.8)	6.53±2.15 5.9 (14.3,3.8)	0.773
1st hour after EST H-	5.93±2.38 5.1 (11.9,1.8)	5.82±1.95 5.6 (12.6,3.7)[b]	0.861
3rd hour after EST	5.93±2.73 5.5 (3.0,14.8)[a]	4.86±1.55 4.3 (8.3,3.0)[c]	0.190
Chi-square*	18.347	38.262	
p*	0.001	0.001	

Data are presented as mean±SD and median (minimum, maximum) values
*-repeated measurements Friedman test for trend
Between time paired comparisons - Wilcoxon test with Bonferroni correction: [a]p=0.024 for baseline vs 3rd hour after EST in control group, [b]p=0.003 for baseline vs 1st hour after EST in the CAD group, [c]p<0.0001 for baseline vs. 3rd hour after EST in CAD group
#Mann-Whitney U test for between groups comparisons
CAD - coronary artery disease, EST - exercise testing, H-FABP-heart-type fatty acid binding protein

Table 4. Comparison of the differences in H-FABP levels between groups

HAFBP, ng/ml	Control group (n=19)	CAD group (n=26)	p#
Difference 1 (Peak EST vs basal)	0.052±2.459 (-2.479/-0.355)	1.114±2.229 (-2.508/-0.384)	0.138
Difference 2 (1st hour after EST vs. basal)	0.836±2.177 (-2.384/-0.400)	1.829±2.363 (-2.369/-0.384)	0.158
Difference 3 (3rd hour after EST vs. basal)	0.837±2.070 (-3.397/-0.508)	2.790±2.569 (-3.349/-0.555)	0.009

Data are presented as mean±SD, #Mann-Whitney U test
CAD - coronary artery disease, EST - exercise stress testing, H-FABP - heart-type fatty acid binding protein

Although the idea that the deterioration of cell membrane integrity, which is the most prominent characteristic of irreversible cell injury, needs to occur to release enzymes and macromolecules like cTn from ischemic myocardium is common, previous studies have shown that temporary ischemia might change cell membrane permeability (21). Askenasy et al. (22) demonstrated

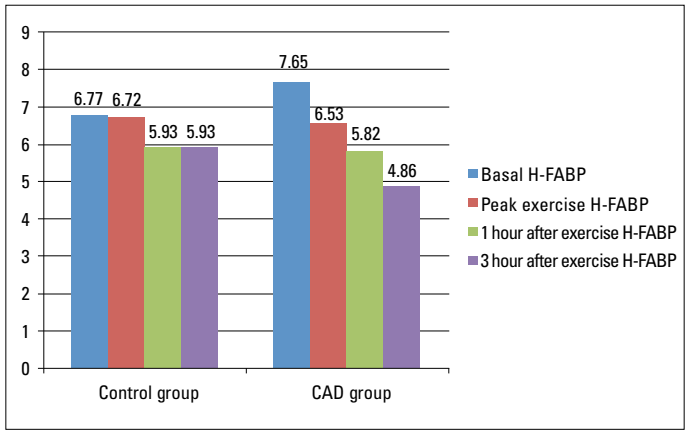

Figure 1. Changes of H-FABP levels during and after exercise testing
CAD - coronary artery disease, H-FABP - heart-type fatty acid binding protein

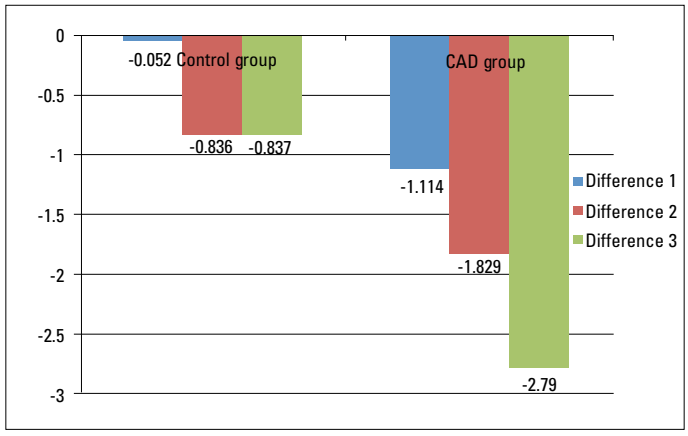

Figure 2. Comparison of the differences in H-PABP measurements between groups
CAD - coronary artery disease, H-FABP - heart-type fatty acid binding protein

that cell membrane permeability against macromolecules following reperfusion increased in association with ischemia duration. While the molecular weight of CK-MB is 86 kDa and TnT 33 kDa, H-FABP is 15 kDa, a much lower molecular weight than CK-MB and cTns, all of which are myocardial injury markers (23). Therefore, we thought that the membrane permeability increase resulting from ischemia might especially cause H-FABP molecules to pass through the cell membrane because these molecules exist freely and abundantly in the myocardium cell cytoplasm and because H-FABP molecules have a lower molecular weight than the other myocardial injury markers. As a result, this condition may lead to an enhancement in the plasma level of H-FABP.

Previous studies have indicated that H-FABP is not only an early and sensitive marker for detecting myocardial damage in ACS (4-8) but also high in patients with ACS whose cTnT levels were normal 12 hour after the onset of symptoms (24). It was also noted that membrane permeability increases due to severe myocardial ischemia might cause H-FABP molecules, ample in cytoplasm, to be released into plasma (24). In a study conducted by Tambara et al. (25), 17 patients waiting for coronary artery bypass grafting operations who had experienced ischemic symptoms and/or ECG changes within last 24 hour were compared to the other 17 patients, who had no ECG changes and/or ischemic symptoms in terms of H-FABP, TnT, and CK-MB levels in the pericardial liquid or in the serum samples obtained after the thoracotomy procedure. In addition, Tambara et al. (25) investigated whether the release of H-FABP into the circulation and pericardial liquid occurred as a response to myocardial ischemia. In that study, they demonstrated that there was no difference between two groups for CK-MB and cTnT levels in serum and pericardial liquid. Although the serum H-FABP levels in both groups were slightly over the normal range, there was no statistical difference. On the other hand, H-FABP levels in pericardial liquid were found to be higher in patients who had experienced ischemic symptoms and/or ECG changes within last 24 hour. That study suggests that H-FABP levels in pericardial liquid could reflect myocardial ischemia within the last 24 hour and might increase in cases of not only myocardial infarction but also myocardial injury related to myocardial ischemia.

The patients in the above studies all have ACS. However, studies evaluating whether myocardial ischemia in patients with stable angina may affect H-FABP levels are quite rare. There is only one study designed to evaluate this situation in the literature (13). In that study, coronary angiography was performed on 47 patients with stable angina after EST. cTnI, H-FABP, and glycogen phosphorylase BB isoenzyme levels were measured in all patients prior to EST and after EST at 3 hour and 5 hour. When the results of this study were compared between 33 patients with CAD and 14 control patients with normal coronary anatomy, no increases were detected with regard to the measurements before EST as compared to the values after EST for all the markers studied, and H-FABP levels were also found to be unchanged. Ischemia induced by EST was observed in 23 of 33 patients from the CAD group; however, there was no difference and no increases detected again in respect to H-FABP levels for both groups when these patients were compared to the other 10 patients without ischemia during EST. It is well known that H-FABP level starts to increase in the first hour of ACS, but in this study, H-FABP levels were evaluated at 3 hour and 5 hour after EST. If temporary myocardial ischemia during EST cannot elevate H-FABP levels as much as in myocardial infarction and removal of these molecules from plasma occurs quickly by the kidneys, detection of increases in this marker may be more effective if the measurements are made in early hours.

In our study, designed after taking into consideration all these findings, in 26 patients with CAD and 19 with normal coronary anatomy, H-FABP levels were measured from the samples obtained at the basal level (prior to EST), peak exercise level (after 10 min of exercise), and at 1h and 3 h after EST. None of the measurements differed between the two groups. The means of the H-FABP values measured at particular intervals for each group tended to decline from the basal level to the 3rd hour level. When comparing the peak exercise measurement to the basal level (difference 1), the 1 hour measurement to the basal level (difference 2), and the 3 hour measurement to the basal level

(difference 3) in each group, differences 2 and 3 in the CAD group were statistically significant (p=0.003 and p<0.0001, respectively). However, only difference 3 in the control group reached statistical significance (p=0.024). Difference 3 measured in the CAD group was higher than in the control group (p=0.009). In other words, a statistically significant decrease at the 3rd hour measurement compared to the basal level in the CAD group was more apparent than in the control group. As a result, H-FABP levels after EST did not change in all patients. Contrary to expectation, H-FABP levels were inclined to decrease. In addition, there was no difference in respect to the all blood samples taken before and after EST when H-FABP levels were compared between the groups that had ischemia during EST and those that did not. These results indicate that H-FABP levels do not increase because of myocardial ischemia occurring during EST.

The decrease in H-FABP levels that occurred in opposition to our expectation likely resulted from exercise-induced proteinuria. Exercise-induced proteinuria is a well-known condition that is thought to be caused by increased glomerular permeability and the exceeding of tubular reabsorbtion capacity (26, 27). The proteinuria that occurred after mild exercise was predominantly of the glomerular type (26). Proteins with low molecular weight (<20 kDa) can easily cross the glomerular capillary membrane and were substantially reabsorbed by proximal tubules (28). It was demonstrated in healthy persons who had undergone exercise testing with the standard Bruce protocol that the albumin-creatinine ratio in urine samples taken within 15 min after exercise was increased and reached the level of microalbuminuria. However, it was found that this ratio returned to its normal level in urine samples taken after 24 h (29).

It was demonstrated that in healthy individuals oxidants, such as peroxide, were increased after treadmill EST and that the balance was shifted toward oxidative stress with a reduction in total antioxidant capacity (30). Exercise related to oxidative stress was said to be a contributory factor to incidence of post-exercise proteinuria (31, 32). Therefore, levels of H-FABP with a molecular weight of 15kDa were likely significantly decreased in both groups due to exercise-induced proteinuria.

It is known that hyperlipidemia, hypertension, diabetes, smoking, and aging, which increase oxidative stress, lipid peroxidation, and the production of reactive oxygen radicals, are related to the mechanism of atherosclerosis formation (33). In our study, the reason for the significant decrease in H-FABP levels at the 3 h measurement in the CAD group, which was more apparent than in the control group, may be the abovementioned oxidative stress increase related to the pathogenesis of atherosclerosis or the occurrence of more proteinuria associated with the increase of oxidative stress due to myocardial ischemia.

Proteinuria is a characteristic of diabetic nephropathy. A study evaluating exercise-induced proteinuria in diabetic patients demonstrated that diabetics had more prominent proteinuria (as measured at the basal level) than healthy individuals (34). The number of diabetic patients in the CAD group in our study is considerably higher (11 patients in the CAD group and 2 in the control group, or 42.3% and 10.5%, respectively). This might be the reason for the greater decrease in H-FABP levels measured at the 3 h after EST in our CAD group. However, further studies are needed to determine the absolute mechanism and the clinical importance of this decrease.

Skeletal muscle has smaller levels of H-FABP, as compared to heart muscle (35). Consequently, situations leading to muscle damage, such as cardioversion, multiorgan insufficiency, and heavy exercise, may also increase H-FABP levels. Therefore, the usage of H-FABP as a marker of myocardial damage is problematic (36). The myoglobin/H-FABP ratio in plasma could be used as a solution to this problem (37, 38). EST is not a kind of heavy exercise. Hence, as seen in our study, a decrease in H-FABP levels after exercise like EST may lead to false negatives results in patients who are suspected of having ACS.

Study limitations

The major limitations of this study were the small sample size, and we did not measure H-FABP levels in urine samples at the same time periods. We refrained from doing so, as we did not think H-FABP levels would decrease after EST with exercise-induced proteinuria as a possible mechanism.

Conclusion

Our study indicates that H-FABP levels do not increase because of myocardial ischemia occurring during EST and so it can not improve the diagnostic performance of EST in combination with EST data. The decrease in H-FABP levels that occurred in opposition to our expectation likely resulted from exercise-induced proteinuria. And also these results can explain false negative results obtained from the patients who were suspected of having ACS and myocardial damage situations by abovementioned mechanism. However, more detailed studies are needed to investigate this subject.

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Conflict of interest: None declared.

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