Original paper

Exercise h-FABP plasma concentration in healthy subjects

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Abstract

Introduction: Heart fatty acid binding protein (h-FABP) is considered a heart specific marker of necrosis, but it is also present in the skeletal muscles. The aim of the study is to check whether h-FABP plasma concentration increases in healthy subjects after exercise.
Material and methods: Plasma h-FABP concentrations were measured in 16 healthy men and 3 healthy women (mean age 24) before and after their participation in the 11 kilometer-long marathon – XVI Independent Race in Warsaw by means of a point-of-care semi-quantitative test.
Results: All 19 runners had a negative result of the pre-run h-FABP test (<7 µg/l). Out of eighteen runners who finished the race: 9 (50%) presented with a positive result of the point-of-care test, 7 runners (39%) had a negative result and in 2 participants the test showed invalid results. All studied subjects were free of cardiologic risk factors, recent infections and were not on any medication. All positive results were found among the male subjects.
Discussion: Plasma h-FABP can increase after moderate exercise probably due to skeletal muscle injury. The concentrations observed after moderate exercise are generally lower than those reported for the acute myocardial infarction.
Conclusion: The point-of-care h-FABP test might be therefore useful in the diagnosis of myocardial necrosis in subjects with angina pectoris and exercise induced chest pain.

Key words: fatty acid binding proteins, bedside testing, muscular stress.

Introduction

Heart fatty acid binding protein (h-FABP) is considered a heart specific protein which binds fatty acids (FABP) within the cardiac muscle cells [1]. It has been reported as an early marker of myocardial ischemia with sensitivity >80% in detecting acute myocardial infarction (AMI) within 30-210 minutes after the symptom onset [2]. It usually peaks after 6h and returns to baseline values after 24h. Although the cardiac tissue contains a 0.5 mg/g wet weight of the h-FABP, the protein is also present in the skeletal muscles in concentrations varying between 0.05 and 0.2 mg/g wet weight of tissue [3, 4].

Physical exercise may influence plasma concentrations of biochemical markers in healthy subjects. In one study the h-FABP concentrations were measured using the quantity one-step immunosorbent assay (sandwich ELISA) in six healthy men after 20 minutes of down-hill running. Mean h-FABP concentrations were significantly elevated beginning from 30 minutes after running (13.9 fold increase of the initial mean concentration 3 µg/l) [5]. In another study h-FABP concentrations were measured after acute and

chronic exercise in a group of 14 junior rowers using the same ELISA sandwich method [6]. The percentage increase in h-FABP immediately after training was 78% after a time trial for 4 km and up to 362% after a steady state rowing for 12-14 km followed by weight training. When the chronic effect of exercise was studied, the baseline h-FABP levels were independent of previous training.

The normal ranges of the h-FABP plasma concentrations vary between 0 and 5 µg/l [7, 8]. The cut-off point for h-FABP in detecting AMI within 6 hours of the onset of chest pain with almost 81.8% sensitivity and 86.4% specificity was reported to be >12 µg/l [9]. Recently a new point-of-care test for measuring of h-FABP with a cut-off point of 7 µg/l was introduced. Knowing that the h-FABP plasma concentrations increase after an exercise and that the point-of-care test cut-off points are lower than those reported for the AMI we wanted to study whether we would obtain positive results of the bedside test in healthy subjects after exercise.

Material and methods

The study included healthy subjects which took part in a 11 kilometer-long marathon – XVI Independence Race, 11th November 2004, Warsaw, Poland. H-FABP concentrations were assessed in each subject before the start of the race and after the end of run. Additional data concerning the prevalence of common cardiologic risk factors was collected, including: hypertension, hyperlipidemia, positive family history of cardiovascular incidents, cigarette smoking, body mass index (BMI) and cholesterol-rich diet. All subjects were asked about recent infections, drug intake, other chronic diseases and the number of hours of training per week. The baseline characteristics of the studied population are shown in Table I.

Table I. Baseline characteristics of the studied population

Characteristics	Description
Age	mean 24 (range 17-31)
Male sex	16/19
Mean BMI*	22.3
History of cigarette smoking	4/19
Blood pressure (mmHg)	mean 122/79
Hyperlipidemia	0/19
Cholesterol-rich diet	1/19
Positive family history of cardiovascular diseases	1/19
Chronic diseases**	1/19
Recent infections	0/19
Any medications	0/19

**BMI – body mass index*
***allergy towards dust*

The test execution was performed with blood samples (3-4 drops) taken from the fingertip by means of the automatic lancet and applied into the funnel according to manufacturer instructions. Before sampling the participant's fingertip was cleaned with 75% alcohol and dried before the puncturing and blood taking performed by qualified personnel. Test results were read after 15 minutes independently by two persons. Before the participation in the study all runners signed an informed consent.

H-FABP plasma concentrations were determined using a CardioDetectMed point-of-care semi-quantitive test (rennesens, Germany) [10]. It is an immunochromatographic lateral flow test and consists of a nitrocellulose membrane which is labeled with special antibodies on two lines. The so-called „Result" line is coated with non-labeled monoclonal anti-h-FABP antibodies. The "Control" line is coated with non-specific anti-mouse IgG. The blood sample applied into the funnel passes through a blood separator and runs through a conjugate pad with gold-labeled monoclonal h-FABP antibodies. Any h-FABP are bound to these antibodies resulting in an antibody protein complex which as it runs is taken up by the anti-h-FABP antibodies in the "Result" position it becomes visible as a red-purple line. The threshold value of the test is 7 µg/l, but a semi-quantitative estimation of h-FABP concentration is possible according to red-purple line intensity resulting in cut-off points of 7, 15, 25 and 100 µg/l.

Results

The studied population consisted of 19 healthy subjects (16 men and 3 women) aged 17-31 (mean age 24). The mean time of the race achieved by the described runners was 50 minutes (range 38-69 min). The mean time between the end of the run and blood sampling was 29 minutes (range 15-50). Examples of positive, negative and invalid test results are shown in Figure 1.

All 19 runners had a negative result of the pre-run h-FABP test (<7 µg/l). Out of 18 runners who finished the race: 9 (50%) presented a positive result of the point-of-care test, 7 runners disclosed a negative

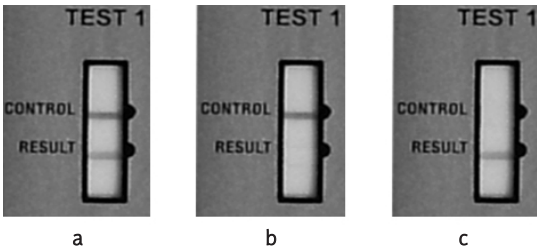

Figure 1. Bedside h-FABP test interpretation: a. positive result, b. negative result, c. invalid result – only a „Result" line is visible or no lines are visible (not shown)

Table II. Characteristics of the groups with increased and normal h-FABP plasma concentrations

	Increased FABP	Normal FABP
Number of subjects	9	7
Male sex*	9/9	4/7
BMI**	22.2	22.1
Time of run (minutes)	52	50
Time-to-test*** (minutes)	79	83
Training (hours/week)	6.5	7

**Two-sided Fisher's exact test p=0.06*
***BMI – body mass index*
****Time-to-test is the time between the start of the race and the execution of the second h-FABP test*

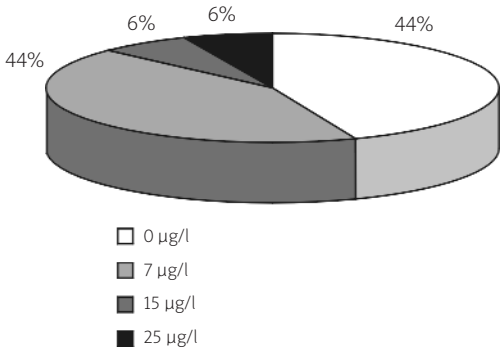

Figure 2. The change in h-FABP plasma concentrations after a 11 km run in the studied population

result (39%), while two tests showed invalid results. None of the runners reported chest pain or any chest discomfort during and after the race. Out of 9 subjects with positive results of the second test in 7 runners h-FABP increased up to 7 µg/ml, in 1 runner more significantly up to 15 and in 1 up to 25 µg/l. No increase up to 100 µg/l was observed in any of the samples. Changes in plasma h-FABP levels after the exercise are shown in Figure 2.

All positive results were found only among the male subjects (p=0.06).

There was no other significant difference in baseline characteristics between the groups according to plasma h-FABP. The obtained results were independent of the time of the run, time till the execution of the second test and hours of training per week (p=NS for all). The characteristics of the groups with increased and normal h-FABP plasma concentrations after the race are presented in Table II.

Discussion

The H-FABP concentration increased on exercise in healthy subjects independently of the time of the run, time-to-test and hours of training per week. Since none of the runners suffered from heart disease or reported any chest pain or discomfort we postulate the skeletal muscle origin of the h-FABP plasma elevation. In one of the previous studies, when four biochemical markers were examined only h-FABP, myoglobin (Mb), and creatinine kinase (CK) increased in plasma after exercise while cardiac troponin I (cTnI) was not detectable in the investigated athletes [5]. High cardiac specificity of cTnI supports the hypothesis of the skeletal muscle source of the h-FABP elevation after exercise [11].

Lower plasma concentrations of h-FABP at rest in healthy women comparing to men had been also reported [12] which might have caused an undetectable increase in the h-FABP levels in the studied female runners in our group.

The present study for the first time assessed the validity of the point-of-care h-FABP test among young healthy subjects. The exercise achieved by runners (about 13 METS) is far more extensive than efforts performed by patients with angina pectoris presenting with chest pain. On the other hand, the h-FABP plasma concentrations observed in the studied runners were in most cases below the levels characteristic of the AMI.

Two cases in which the h-FABP test disclosed an invalid result were caused by the difficulties in blood collection probably due to peripheral vessels constriction resulting in an insufficient sample volume applied into the funnel. One limitation of this study is a relatively small group of the studied subjects and therefore should be confirmed on larger populations.

The other limitation consists of the imprecise data concerning hyperlipidemia which presence should be excluded by detection of plasma cholesterol concentrations. We also did not compare changes in the h-FABP plasma concentrations with the plasma levels of other biochemical markers of the heart-muscle injury (Mb, CK, cTnI).

Conclusion

The H-FABP point-of-care test is a good marker for distinction of the skeletal and cardiac muscle injury in patients with angina pectoris and exercise induced chest pain.

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References

1. Glatz JF, van der Vusse GJ, Simoons ML, Kragten JA, van Dieijen-Visser MP, et al. Fatty acid-binding protein and the early detection of acute myocardial infarction. Clin Chim Acta 1998; 272: 87-92.

2. Kleine AH, Glatz JF, Van Nieuwenhoven FA, Van der Vusse GJ. Release of heart fatty acid-binding protein into plasma after acute myocardial infarction in man. Mol Cell Biochem 1992; 116: 155-62.
3. Alhadi HA, Fox KA. Do we need additional markers of myocyte necrosis: the potential value of heart fatty-acid-binding protein. QJM 2004; 97: 187-98.
4. Yoshimoto K, Tanaka T, Somiya K, Tsuji R, Okamoto F, et al. Human heart-type cytoplasmic fatty acid-binding protein as an indicator of acute myocardial infarction. Heart Vessels 1995; 10: 304-9.
5. Sorichter S, Mair J, Koller A, Pelsers MM, Puschendorf B, Glatz JF. Early assessment of exercise induced skeletal muscle injury using plasma fatty acid binding protein. Br J Sports Med 1998; 32: 121-4.
6. Yuan Y, Kwong AW, Kaptein WA, Fong C, Tse M, et al. The responses of fatty acid-binding protein and creatine kinase to acute and chronic exercise in junior rowers. Res Q Exerc Sport 2003; 74: 277-83.
7. Wodzig KW, Pelsers MM, van der Vusse GJ, Roos W, Glatz JF. One-step enzyme-linked immunosorbent assay (ELISA) for plasma fatty acid-binding protein. Ann Clin Biochem 1997; 34: 263-8.
8. Tsuji R, Tanaka T, Sohmiya K, Hirota Y, Yoshimoto K, et al. Human heart-type cytoplasmic fatty acid-binding protein in serum and urine during hyperacute myocardial infarction. Int J Cardiol 1993; 41: 209-17.
9. Ishii J, Wang JH, Naruse H, Taga S, Kinoshita M, 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; 43: 1372-8.
10. Chan C, Cheng WS, Glatz JF, et al. Early diagnosis of acute myocardial infarction using immunosensors and immunotests. Analytical Letters 2003; 36: 1987-2004.
11. Adams JE 3rd, Bodor GS, Davila-Roman VG, Delmez JA, Apple FS, et al. Cardiac troponin I. A marker with high specificity for cardiac injury. Circulation 1993; 88: 101-6.
12. Pelsers MM, Chapelle JP, Knapen M, Vermeer C, Muijtjens AM, et al. Influence of age and sex and day-to-day and within-day biological variation on plasma concentrations of fatty acid-binding protein and myoglobin in healthy subjects. Clin Chem 1999; 45: 441-3.