

Direct comparison of the diagnostic value of point-of-care tests detecting heart-type fatty acid binding protein or glycogen phosphorylase isoenzyme BB in patients with acute coronary syndromes with persistent ST-segment elevation

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Abstract

Background: Myocardial infarction (MI) with its complications is one of the most serious challenges in contemporary cardiology. Among biochemical markers of myocardial necrosis, heart-type specific fatty acid binding protein (h-FABP) showed excellent sensitivity and specificity for the early diagnosis of an acute MI. The h-FABP is released rapidly (after 30 min) from the cardiomyocyte to the circulation in response to myocardial injury and may be useful for rapid confirmation or exclusion of MI. In recent years, glycogen phosphorylase BB (GP-BB) also emerged as a promising early specific marker of myocardial necrosis. Rapid, qualitative “point of care” tests (POCT) detecting h-FABP (Cardio Detect med) and GP-BB (Diacordon) have recently become available.

Aim: To evaluate and compare qualitative POCTs detecting h-FABP and GP-BB in patients with an acute coronary syndrome (ACS).

Methods: We studied 52 patients with a strong suspicion of ACS with persistent ST-segment elevation and chest pain lasting less than 6 hours. The ultimate diagnosis of ST-segment elevation MI (STEMI) was confirmed in case of a second (6 h after admission) positive quantitative result of a cardiac troponin T (cTnT) test. On admission, POCTs to detect both h-FABP and GP-BB were performed. The study population was divided into two groups, with chest pain lasting < 3 h (n = 20) or 4–6 h (n = 32). All patients underwent coronary angiography and angioplasty if indicated. The sensitivity of the analysed biomarkers of myocardial necrosis was calculated.

Results: The sensitivity of h-FABP (84%) was superior in comparison to the other biomarkers, GP-BB and cTnT, which had sensitivity of 64% and 50%, respectively. Comparison of typical parameters of the diagnostic value of a test (sensitivity, predictive values and accuracy) in both time periods demonstrated that h-FABP was superior to GP-BB. In particular, sensitivity and accuracy of h-FABP was excellent in the group of patients with chest pain lasting < 3 h, with sensitivity of 79% for h-FABP and only 47% for GP-BB. Sensitivity and accuracy of cTnT were significantly lower (32% and 35%, respectively).

Conclusions: The h-FABP seems to be an excellent early biomarker of cardiac necrosis in the group of patients with chest pain lasting < 3 h. The GP-BB can be also used as a biomarker of myocardial necrosis, but its sensitivity in the early phase of MI is limited.

Key words: acute coronary syndrome, myocardial infarction, markers of necrosis, h-FABP, GP-BB

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INTRODUCTION

Biochemical diagnosis of acute coronary syndromes (ACS), and particularly myocardial infarction (MI), is one of the most rapidly developing areas of the 21st century cardiology. This results from the fact that MI with its complications, including sudden cardiac death, is one of the most serious challenges in contemporary clinical cardiology. Extrapolation of data from the Polixh registry of ACS [1] to the general Polish population gives more than 140,000 hospitalisations due to ACS annually, i.e. 4000 per 1 million inhabitants, including nearly 60,000 ST-segment elevation MI (STEMI).

Advances in new cardiac biomarkers resulted in an updated universal definition of MI (2007), based on more sensitive and specific biological markers of myocardial necrosis compared to those used previously. Cardiac troponins, despite unquestionable role in the diagnosis of an acute MI (gold standard), have major drawbacks. First, due to kinetics of troponin release from the area of myocardial necrosis, troponin is not an early marker, as it can be detected in peripheral blood only after 5–6 hours. Therefore, especially in STEMI with chest pain lasting < 5 hours, the result of the troponin test (often initially negative) has no role in instituting reperfusion therapy which should be started as soon as possible, regardless of the initial results of biomarker testing to detect myocardial necrosis. Second, troponins are biochemical evidence of myocardial necrosis but not necessarily of ischaemia of vascular origin, and they may be released in many other conditions leading to myocardial necrosis. Thus, there is a continuous search for novel markers of infarction that would show superior sensitivity and specificity especially in the first hours and decrease diagnostic uncertainty in this period, using so called two-marker strategy. Of note, growing use of rapid biochemical tests that can often be used at the bedside (so called point of care tests — POCT) resulted in a significant reduction of clinical uncertainty when the ECG findings are equivocal. Among novel biochemical markers of myocardial necrosis, heart-type specific fatty acid binding protein (h-FABP) and glycogen phosphorylase isoenzyme BB (GP-BB) are of particular interest.

The h-FABP belongs to a family of small (12–15 kDa) cytoplasmic proteins. These proteins are present in tissues metabolising free fatty acids in many mammals, participating in fatty acid transport, and show some tissue specificity, with highest levels noted in the heart and the liver. The h-FABP is a 132-aminoacid protein, present in the cytoplasm of mainly cardiomyocytes (at the level of about 0.5 mg/g tissue) and skeletal myocytes (at the level that is approximately 10 times lower than in cardiomyocytes) [2].

In healthy subjects, plasma h-FABP concentration is very low (range 1–11.4 µg/L; median 1.5 µg/L) [3]. An increase in h-FABP level above a defined cutoff value may occur already approximately 0.5 h after the onset of chest pain. Maximum level was seen at 4 h in patients undergoing reperfusion therapy [4] and after 8 h without reperfusion therapy [5], with

normalisation within 24 h [6]. A qualitative rapid immunochemical POCT (CardioDetect med) has been available for some years that may give reliable evidence of myocardial necrosis already 30 min after the onset of chest pain, i.e. within so called “golden hour” [7, 8]. This kinetics is the main advantage of h-FABP, despite lack of full cardiac specificity, as this test allows detection of necrosis when other plasma biomarkers, including troponins, are still negative.

Recently, a POCT detecting another early marker of cardiomyocyte damage, GP-BB, has been introduced in patients with ACS. Glycogen phosphorylase is a key enzyme taking part in glycogenolysis. Three isoenzymes exist, known as BB (brain and heart), MM (muscle) and LL (liver), that have different function and immunologic properties [9, 10]. The GP-BB is associated with glycogen in the sarcoplasmic reticulum (mainly in cardiomyocytes) and catalyses the initial step of glycogenolysis, i.e. release of glucose-1-phosphate from glycogen. Glycogen phosphorylase from damaged myocardial cells may indicate initiation of glycogenolysis resulting from acute myocardial ischaemia. Low oxygen level in the myocardium induces glycogenolysis leading to loss of plasma membrane integrity and extracellular leak of soluble intracellular proteins. This process is accompanied by a rapid increase in serum GP-BB level in patients with an acute MI, prior to the rise of other biomarkers such as creatinine kinase, CK-MB, myoglobin and troponin T [11]. Initial studies comparing this biomarker with established cardiac markers showed increased GP-BB level within 4 h from the symptom onset [12]. The GP-BB level in patients with type I infarction usually peaks between 6 and 20 h from the symptom onset and normalises within 1–2 days after MI [13]. It is still not clear whether GP-BB release indicates irreversible cardiomyocyte damage or may also occur in severe ischaemia and some loss of integrity of cardiomyocyte cellular membrane.

The aim of the study was to perform a direct comparison of the diagnostic value of h-FABP and GP-BB in a group of 52 patients with an ACS with persistent ST-segment elevation and chest pain lasting < 6 hours.

METHODS

Study group

We studied 52 consecutive patients with suspicion of ACS with persistent ST-segment elevation who were hospitalised in our department from January to September 2009.

Inclusion criteria were the following: (1) ACS within 6 h from the symptom onset and persistent ST-segment elevation in the baseline ECG; (2) signed informed consent, and (3) lack of exclusion criteria (ACS without persistent ST-segment elevation, or unclear timing of symptoms).

Among included patients, second troponin T measurement at 6 h after admission allowed the diagnosis of STEMI type I according to the universal definition of MI in 50 patients (in 1 patient, patent coronary arteries were found despite suggestive ECG changes, and 1 patient was finally diagnosed with variant angina). The “gold standard” confirming myocardial

Table 1. Basic demographic, clinical and angiographic data and main risk factors in the study group

Variable	Chest pain < 3 h (n = 20)	Chest pain 4–6 h (n = 32)	P
Number of patients	20	32	
Age [years]	60.5 (60.9 ± 10.7)	60.0 (60.9 ± 9.6)	NS
Body mass [kg]	82.5 (81.9 ± 11.8)	77.0 (79.5 ± 18.1)	NS
BMI [kg/m ²]	27.9 (28.4 ± 3.4)	26.7 (27.3 ± 4.8)	NS
Women/men	40%/60%	25%/75%	NS
Hypertension	95%	56%	0.01
History of IHD	40%	38%	NS
Smokers	40%	50%	NS
Previous MI	20%	19%	NS
Infarct-related artery:*			
LAD	53%	55%	NS
LCx	21%	26%	NS
RCA	26%	19%	NS

*In both groups in patients with an acute MI only (n = 19 in the early presenting group, n = 31 in the late presenting group); Mann-Whitney test for continuous variables, χ^2 test for categorical variables; BMI — body mass index; IHD — ischaemic heart disease; MI — myocardial infarction; LAD — left anterior descending artery; LCx — left circumflex artery; RCA — right coronary artery

necrosis was elevated troponin T level (> 0.10 ng/mL) at 6 h after admission, with typical dynamics in serial testing, measured quantitatively in our hospital laboratory. All patients underwent urgent coronary angiography and primary angioplasty if indicated, according to the standard of care for patients with an acute MI. For further analysis, the study group was divided into two groups depending on the time from the symptom onset to admission: with chest pain lasting < 3 h (n = 20, including 19 patients with STEMI) or 4–6 h (n = 32, including 31 patients with STEMI). In all patients, blood samples were collected in the emergency room for the purpose of h-FABP and GP-BB testing.

The study was approved by the local ethics committee at the Medical University of Łódź.

Methodology of h-FABP and GP-BB testing

The CardioDetect med test detecting h-FABP is based on immunochemical determination of monoclonal antibodies characteristic for h-FABP, one of them labelled with gold. A few drops of full blood are placed in the reference area of the test card, and the result is read after 10–15 min. The test is positive when two colour stripes are seen in the read area, indicating blood biomarker level of > 7 ng/mL. Blood removes gold-labelled antibodies against h-FABP from the matrix, forming h-FABP-anti-h-FABP complexes that pass the read area. In the test window, these complexes form another, visible complexes with the second antibody (purple stripe on the test card). The Diacordon test detecting GP-BB is based on similar technology using monoclonal antibodies, but the determination is technically more complex, requiring addition of a few drops of a special buffer to the blood sample. Blood sample is then placed on the test card and the test result is read in a similar way after 15 min.

Statistical analysis

Analysis of the statistical parameters was performed using the following licensed software: MedCalc 8.0 for Windows (Frank Schoonjans, 1993–2005) and Statistica 8.0 PL (StatSoft). Empirical data were analysed using basic descriptive statistical methods (mean, median, SD, structure index) and diagnostic tests (sensitivity and accuracy). We used nonparametric χ^2 test for the statistical analysis of categorical variables, the McNemara test for dependent variables, and the Mann-Whitney test for independent variables. A p value < 0.05 was considered statistically significant.

RESULTS

Demographic characteristics, prevalence of risk factors, and distribution of the infarct-related artery are shown in Table 1. Left anterior descending artery was the most common infarct-related artery in both subgroups.

Figure 1 shows sensitivity of the tested biomarkers on admission. Overall, h-FABP was the most sensitive marker among all patients with STEMI. Table 2 shows sensitivity, positive predictive value and diagnostic accuracy of the tested biomarkers. In both time periods, h-FABP was a better diagnostic marker than GP-BB, with significantly higher sensitivity and accuracy of the test detecting h-FABP within 3 h from the onset of chest pain. In the first time period, sensitivity and accuracy of cardiac troponin was poor.

DISCUSSION

In this study we attempted to determine which of the earliest myocardial necrosis markers available as commercial POCTs has superior diagnostic value during the first hours of evolving MI. This is a major clinical problem if we consider the importance of earliest possible diagnosis of myocardial ischa-

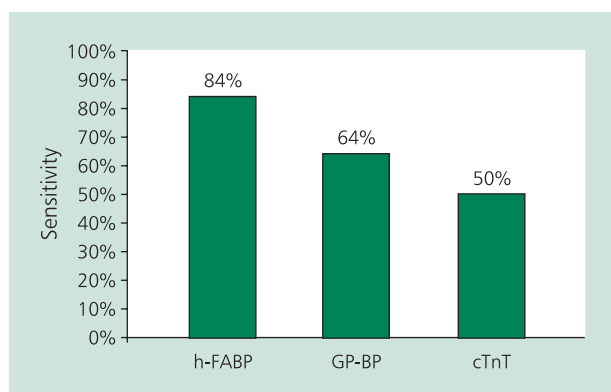


Figure 1. Sensitivity of heart-type fatty acid binding protein (h-FABP), glycogen phosphorylase isoenzyme BB (GP-BB) and cardiac troponin T (cTnT) in detecting STEMI in the whole study group

emia and necrosis in patients with ACS with persistent ST-segment elevation. We still need to optimise our diagnostic approach, including prehospital diagnosis in patients with STEMI, in order to allow rapid institution of the reperfusion therapy. Despite many recent logistic and technical efforts, the latter is still often initiated with excessive delay, as evidenced by a larger proportion of patients in our study who presented > 3 h from the symptom onset.

Our findings in regard to sensitivity, positive predictive value and accuracy of the tested biomarkers showed that h-FABP was a more useful early marker of STEMI, than GP-BB and troponin I. Superiority of h-FABP was even more evident in patients presenting within 3 h from the symptom onset. In this subgroup, h-FABP was more sensitive than GP-BB and troponin. Thus, only h-FABP was a highly sensitive and accurate biomarker, while GP-BB was only modestly better than troponin which was, as expected due to its kinetics, least sensitive and

accurate. Our findings show that only h-FABP may be considered a really useful early biomarker of myocardial necrosis.

During the last few years, there has been growing evidence in the literature confirming the usefulness of h-FABP in the early diagnosis of MI. Okamoto et al. [14] evaluated the usefulness of h-FABP testing in a group of nearly 200 patients with chest pain lasting less than 12 h. Overall sensitivity of h-FABP was 92.9%, compared with 88.6% for myoglobin and 18.6% for CK-MB. Suzuki et al. [15] reported that sensitivity of h-FABP and cardiac troponin T was 59.1% and 15.4%, respectively, if measured within 3 h from the onset of pain, and 65.2% and 56.4%, respectively, if measured > 3 h. The usefulness of h-FABP testing, particularly to exclude MI, was also examined by Chan et al. [16] who showed that serial h-FABP testing allowed reliable diagnosis of MI in 100% of cases. Importantly, serial negative result gave virtually 0% of false negative results in this group of patients. Similar results, with high negative predictive value (95–99%) of a negative h-FABP test in patients with chest pain, were reported by Pagani et al. [17]. The h-FABP testing allowed earlier and more reliable exclusion of MI compared to myoglobin. Pelters et al. [18] also showed superior sensitivity of h-FABP compared to myoglobin in patients with ACS. In our experience, h-FABP has high diagnostic accuracy also in early non-ST-segment elevation ACS. We studied 100 patients with ACS and chest pain within 12 h and showed that initial h-FABP test was superior to conventional necrosis markers [19]. In a recently published study comparing quantitative measurements of h-FABP and troponin in patients with MI within 4 h from the onset of pain, sensitivity of h-FABP was much higher and comparable to our present findings (86% vs 42% for troponin T) [20].

Data regarding diagnostic usefulness of GP-BB testing in the diagnosis of MI are more limited and mainly directed at more clinical biochemistry issues, and the marker was

Table 2. Comparison of sensitivity, positive predictive value and accuracy of the analysed biomarkers on admission in both subgroups according to the duration of symptoms

Parameters	h-FABP	GP-BB	cTnT
< 3 h from symptom onset (n = 20)			
Sensitivity [%]	79%	47%	32%
Concordance with a reference cTnT test at 6 h	Concordant: $p > 0.05$; test _{McNemara} = 2.25	Discordant: $p < 0.01$; test _{McNemara} = 8.10	Discordant: $p < 0.001$; test _{McNemara} = 8.10
Positive predictive value	100%	100%	100%
Accuracy	80%	50%	35%
4–6 h from symptom onset (n = 32)			
Sensitivity [%]	87%	74%	61%
Concordance with a reference cTnT test at 6 h	Concordant: $p > 0.05$; test _{McNemara} = 2.26	Discordant: $p < 0.05$; test _{McNemara} = 6.13	Discordant: $p < 0.01$; test _{McNemara} = 10.1
Positive predictive value	100%	100%	100%
Accuracy	87%	75%	61%

Discordance in the McNemara test means a significant difference between parameters of a diagnostic value of a given biomarker and the reference test; h-FABP — heart-type fatty acid binding protein; GP-BB — glycogen phosphorylase isoenzyme BB; cTnT — cardiac troponin T

usually measured quantitatively using the ELISA method. We are not aware of any studies that would directly compare h-FABP and GP-BB in STEMI patients, particularly using qualitative POCTs. Peetz et al. [21] showed that, when measured within initial 6 h, GP-BB was more sensitive and specific than myoglobin and CK-MB mass (95.5–100% vs 85–95% and 71.4–91.3%, respectively). Of note, elevated GP-BB level was also found (at 3 h) in 93.9% of patients who received the final diagnosis of unstable angina. Thus, GP-BB may be a marker of ischaemia or infarction and not only established necrosis. Rabitzsch et al. [22] found that GP-BB was most sensitive and superior even to myoglobin, with detectable rise within 2 h from the onset of chest pain. Mair et al. [23] showed that among all biomarkers tested on admission, including CK-MB, CK-MB mass, myoglobin, troponin T and GP-BB, only the latter was significantly increased in the majority of patients with unstable angina. These findings again suggest that GP-BB might share some features of “classical” necrosis markers (such as troponin and CK-MB mass) and specific ischaemia markers (such as ischaemia-modified albumin) and thus its discriminative value in regard to distinguishing necrosis (infarction) and ischaemia without necrosis remains debatable.

McCann et al. [24] directly compared performance of h-FABP and GP-BB in an early phase of ACS. In a prospective study of 664 patients presenting to an emergency room due to chest pain, a panel of necrosis and ischaemia markers was determined, including troponin T, h-FABP, GP-BB, myeloperoxidase, matrix metalloproteinase-9, pregnancy-associated plasma protein A, and soluble CD40 ligand. Among these markers, only h-FABP was as sensitive and specific as troponin T, and was significantly more sensitive in patients presenting within 4 h (73% vs 55%, $p = 0.043$).

Limitations of the study

Study limitations included single-centre, pilot nature of the study, small patient sample, and restriction of the study group to STEMI patients.

CONCLUSIONS

The h-FABP detected with a qualitative rapid POCT is a reliable biomarker of STEMI that is particularly useful in the early phase of MI (< 3 h). The GP-BB detected with a rapid POCT may be used as an additional biomarker of myocardial necrosis but its diagnostic accuracy in the early phase of MI is limited.

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Ocena wartości diagnostycznej szybkich testów typu POC wykrywających sercowe białko wiążące kwasy tłuszczowe lub izoenzym BB fosforylasy glikogenowej u pacjentów z ostrym zespołem wieńcowym z przetrwałym uniesieniem odcinka ST

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Streszczenie

Wstęp: Zawał serca (MI) wraz z powikłaniami stanowi jeden z najpoważniejszych problemów współczesnej kardiologii klinicznej. Spośród biochemicznych markerów martwicy mięśnia sercowego białko sercowe wiążące kwasy tłuszczowe (h-FABP) odznacza się wysoką czułością i swoistością we wczesnej fazie MI. Białko to jest uwalniane bardzo szybko (po ok. 30 min od rozpoczęcia dolegliwości bólowych) z martwiczo zmienionych kardiomiocytów i może być bardzo użyteczne zarówno w przypadku wczesnego potwierdzenia, jak i wykluczenia MI. Niedawno pojawiła się także możliwość oznaczania innego wczesnego markera uszkodzenia kardiomiocytów — izoenzymu BB fosforylasy glikogenowej (GP-BB). Jakościowa ocena tych dwóch markerów jest możliwa dzięki komercyjnym testom typu POC (POCT, *point of care test*) wykrywających jakościowo stężenie badanego markera martwicy we krwi obwodowej.

Cel: Celem badań było porównanie przydatności klinicznej jakościowego oznaczania wczesnych markerów martwicy — h-FABP oraz GP-BB. Grupa pacjentów liczyła 52 osoby z ostrym zespołem wieńcowym (OZW) z przetrwałym uniesieniem odcinka ST i czasem trwania bólów zamostkowych nieprzekraczającym 6 godzin.

Metody: Do badania włączono 52 pacjentów z podejrzeniem OZW z przetrwałym uniesieniem odcinka ST, hospitalizowanych w ośrodku, w którym pracują autorzy. Ostatecznie potwierdzono rozpoznanie MI z uniesieniem odcinka ST u 50 chorych (dodatni wynik troponiny T z zachowaniem dynamiki jej stężenia w drugim pomiarze, tj. po 6 h od przyjęcia). U każdego chorego przy przyjęciu oznaczono metodą jakościową h-FABP (Cardio Detect med) i GP-BB (Diacordon). Badaną grupę podzielono dodatkowo wg kryterium czasu trwania bólów zamostkowych na podgrupę z bólami do 3 godzin (n = 20) i 4–6 godzin (n = 32). Wszyscy pacjenci byli poddani pilnej koronarografii i ewentualnej pierwotnej koronaroplastyce wg obowiązującego standardu postępowania u chorych z OZW.

Wyniki: Porównanie czułości badanych markerów martwicy wykazało najwyższą czułość (84%) dla h-FABP, dla GP-BB — 64%, zaś dla troponiny T — 50%. Analiza parametrów określających wartość diagnostyczną testu (czułość, wartości predykcyjne, dokładność) w analizowanych przedziałach czasowych wykazała przewagę h-FABP nad GP-BB. Czუłość i dokładność testu wykrywającego h-FABP w pierwszych 3 godzinach od wystąpienia bólu w klatce piersiowej wynosiła 79%, natomiast dla GP-BB czułość była równa 47%, a dokładność — 50%. Troponina sercowa w tym samym przedziale czasowym charakteryzowała się względnie niską czułością (32%) i dokładnością (35%).

Wnioski: Oceniane jakościowo h-FABP okazało się najbardziej wiarygodnym diagnostycznie wczesnym markerem MI z uniesieniem odcinka ST z ogólną czułością 84% w odniesieniu do innych badanych markerów martwicy (GP-BB — 64%, troponina T — 50%). Test wykrywający h-FABP udowodnił swoją przydatność także w bardzo wczesnej fazie MI w podgrupie chorych z bólem w klatce piersiowej do 3 godzin, znacznie przewyższając czułością test wykrywający GP-BB (79% v. 47%).

Wnioski: GP-BB może być stosowany jako uzupełniający marker martwicy miokardium, ale jego czułość we wczesnej fazie MI jest ograniczona.

Słowa kluczowe: ostry zespół wieńcowy, zawał serca, markery martwicy, h-FABP, GP-BB

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