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RESEARCH ARTICLE

CHANGES IN HEART TYPE FATTY ACID BINDING PROTEIN (H-FABP) AND CERTAIN BIOCHEMICAL PARAMETERS DURING CHRONIC ARTERY DISEASES

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ABSTRACT

Coronary artery disease (CAD) is one of the leading causes of death and morbidity in our country, which is also true for the world in general as well. CAD generally develops atop atherosclerosis events. In this study, changes in heart-type fatty acid binding protein (H-FABP) and certain other biomarker levels during chronic artery diseases were investigated.. For the present paper, serum samples collected from patients who applied to Van Yüzüncü Yıl University Dursun Odabaş Medical Center Emergency Department and Cardiology Polyclinics with acute ischemic chest pain between January - June of 2019 were evaluated. Patients were not given any drugs or other kinds of substances before sample collection. Of the patients who applied to the cardiology clinic, 24 of these patients were diagnosed with chronic arteritis, 12 patients with cardiological problems were diagnosed with diabetes mellitus (DM), 12 patients with hypertension (HT) problems and heart complaints, and 12 healthy individuals (who were not diagnosed with diabetes, hypertension or CAD) were included as research materials.

Certain markers like Troponin, CK, CK-MB, AST, ALT, LDL-Cholesterol, HDL-Cholesterol, LDH, Glucose, and Creatinine in the blood samples were biochemically determined using an autoanalyzer (Abbott ci16200),while H-FABP values were determined using ELISA method.

As a result of the analyses carried out : LDL-cholesterol values reached their highest in the CAD group (108,06 ± 6,82 MG/DL), while HDL-Cholesterol and LDH values peaked in the CAD+HT group (51,52 ± 3,92 MG/DL), (318,83 ± 37,42 MG/DL), and CK, CK-MB, cTnI, AST, Glucose, creatinine, and H-FABP levels were found to be high in the DM+CAD group. Meanwhile, cTnI

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values were increased in people with HT or DM as well as CAD, but there was no statistical significance. Similarly, LDL-cholesterol levels stayed similar between the groups. Intergroup changes in other biomarkers examined showed the importance of CK and Glucose levels at p≤ 0.001, AST p≤ 0.002, LDH p≤ 0.003, CK-MB p≤ 0.004, HDL-Cholesterol p≤ 0.049, Creatinine p≤ 0.011, and H-FABP p≤ 0.050. H-FABP has recently taken its place in the field of cardiology with increasing importance in the diagnosis of CAD and MI. The findings obtained in this study show that the H-FABP level was increased in all groups except the test group, and we recommend its use as a practical parameter in cardiology clinics.

Keywords: *Diabetes mellitus, Hypertension, H-FABP, Cardiac markers, Coronary artery disease*

1. INTRODUCTION

Coronary artery disease (CAD) is quite prevalent in the world and is associated with serious cardiac effects in many patients. Coronary artery disease is often triggered by physical or psychological stress. Symptoms related to it usually don't last long. That being said, a coronary artery is completely blocked during the disease which can cause permanent damage to the heart muscles (myocardial infarction) if not diagnosed and treated early. This can cause severe symptoms which usually occur in the form of chest pain and shortness of breath. In certain cases, it can result in death. Diabetes, hypertension, smoking, hypercholesterolemia, inactive life, obesity, and genetic factors are possible risk factors for CAD. Rarely, patients with CAD may show no symptoms. Cardiomyocytes are extremely rich in various proteins and enzymes, including troponin, creatine kinase (CK), creatine kinase MB (CK-MB), and lactic dehydrogenase (LDH). These proteins and enzymes can be released and dispersed into the bloodstream following cardiomyocyte necrosis and breakdown. These are then broken down and mixed into the blood and can be used as vital markers for the early diagnosis of coronary artery disease. In addition to these cardiomyocytes Heart-type fatty acid-binding protein (H-FABP), which may be introduced into the blood even earlier, can help facilitate early diagnosis [1].

Although H-FABP is expressed mainly in cardiomyocytes, it also occurs in much lower concentrations in skeletal muscle, renal distal tubular cells, and the brain. There is also the use of H-FABP as a sensitive marker for exercise-induced skeletal injury [2]. Myocardial infarction, congestive heart failure and angina pectoris [3] in the damaged area of the heart, for example, the heart isoform of the first described biochemical marker creatine kinase (CK-MB) has found its place in the clinic as a 'gold standard' protein. [4] Plasma H-FABP concentrations increase and decrease faster than creatine kinase (CK), indicating that H-FABP is more useful than CK for early diagnosis of such damage and monitoring of damage during repeated exercise sessions. The potential use of H-FABP as a rapidly changing biomarker to diagnose the early stages of acute myocardial infarction was studied in [5]. The results of this study were later put to test by many other researchers as well. Normally, plasma or interstitial fluid contains no H-FABP, which is only released following cellular injury to the heart. This release occurs approximately 2 hours following the onset of the symptoms, and studies suggest that it peaks at approximately 4 to 6 hours. Return to its normal baseline occurs within 20 hours. For the next 3 to 4 hours following the symptom onset, H-FABP has more than 80% sensitivity for AMI events. Other heart markers (CK), such as creatine kinase, CK-muscle and brain (MB) (mass or activity), cardiac troponin I (cTnI), and cardiac troponin T (cTnT) will only begin to accumulate in

plasma within 0-6 hours of symptom onset, and their sensitivity has been reported to be around 64% [5].

The main causes of coronary artery disease are the result of the loss of elasticity of the coronary arteries. CAD occurs with lipid plaque accumulation and hardening and rupture of the vessels. These plaques have a high tendency to cause deterioration of the vascular structure and subsequent formation of clots. A significant part of the CAD can result in AMI, and even death if the intervention is late. Most such sudden and untimely deaths are due to atherosclerosis, which often goes unnoticed due to its associated risk factors being prone to modification, prevention, or reduction.

A heart attack occurs due to the death of cells in the heart tissue, which in turn is the result of the blockage of one or more of the main vessels in the heart that feed that particular tissue. This scenario can lead to fatal results. Depending on the severity of the obstruction, symptoms such as sweating, nausea, vomiting, and sometimes fainting may occur, which may be accompanied by severe pain in the chest. Although chest pain indicates a heart attack, it may sometimes be felt in the upper abdomen and stomach area in some people. Such pain is often ignored, thinking that it's just a temporary discomfort caused by the stomach. Generally, in elderly people, the crisis may occur in the form of shortness of breath that increases with effort. These symptoms can be seen in 75-80 out of 100 people. The other 20% occur in a condition called "silent heart attack", which shows no symptoms. The first symptom in these cases is often death [6].

Factors like nutritional disorders, overeating, a fatty diet, consuming too many ready-made foods, hypertension, high cholesterol, coronary arteriosclerosis (Atherosclerosis), diabetes, smoking, obesity, and an inactive life are among the leading causes of heart problems [7].

2. MATERIALS and METHODS

This research was conducted on a total of 60 patients between the ages of 20 and 100, between January 2019 and June 2019, after obtaining permission with the Ethics Committee Approval No: 12.10.2018 decision no 06. Of these 60 individuals, 48 were patients and 12 were healthy.

The relationship between H-FABP and some biochemical and cardiac parameter values (CK-MB, Troponin, AST, ALT, LDL-Cholesterol, HDL-Cholesterol, LDH, Glucose, and Creatinine) of patients admitted to emergency departments of hospitals and cardiology outpatient clinics (with complaints of coronary artery disease and chest pain) were examined.

2.1. Patient Selection

In this study, blood samples collected from patients who applied to Van Yüzüncü Yıl University Dursun Odabaş Medical Center Emergency Department and Cardiology Polyclinics between January 2019 and June 2019 were evaluated. No medication or other substance was given to these people or no application was made. Of the patients who came to the cardiology with similar demographic characteristics, 24 patients (CAD=24) with acute ischemic chest pain and diagnosed with chronic arteritis disease, 12 patients with cardiological problems diagnosed with diabetes mellitus (DM+CAD=12), 12 patients with hypertension problems and again came to the clinic with a heart

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complaint (HT+CAD=12) and 12 healthy people who were not diagnosed with diabetes, hypertension and with similar demographic characteristics CAD (CONTROL=12) as research material it was used. Certain markers like Troponin, CK, CK-MB, AST, ALT, LDL-Cholesterol, HDL-Cholesterol, LDH, Glucose, and Creatinine in the blood samples were biochemically determined using an autoanalyzer (Abbott ci16200), while H-FABP values were determined using ELISA method.

2.1.1. Human heart fatty acid binding protein (H-FABP)

Principle: ELISA Kit Biont, Catalog No: YLA1747HU. This kit uses enzyme-linked immunosorbent assay (ELISA) based on biotin double antibody sandwich technology to assay human heart fatty acid binding protein (H-FABP). H-FABP is added to pre-coated cavities with monoclonal antibodies and then incubated. After that, anti-H-FABP antibodies labeled with biotin are added to combine with streptavidin-HRP, which forms an immune complex. After incubation and washing, unbound enzymes are removed. Substrates A and B are added. Then the solution turns blue, and under the influence of acid, turns yellow. The colorimetric tones of the solution and the concentration of H-FABP are positively correlated.

2.1.2. Assay procedure summary

1. Prepare all reagents, samples and standards.
2. Prepared samples, standards and ELISA solutions are added. They are allowed to react for 60 min at 37 °C.
3. The plate is washed five times. Chromogen solutions A and B are added for color development. Incubate at 37 °C for 10 minutes.
4. The stop solution is added.
5. The OD value is read and calculated within 10 minutes.

Working range： 0.05 ng/ml → 20 ng/ml 25

Sensitivity：0.01 ng/ml

2.1.3. Statistical analysis

For the descriptive statistics for the features emphasized, the average is expressed as the SEM value. The Kruskal-Wallis test was used to compare the groups in terms of these features. In the calculations, the statistical significance level was taken as 5%. SPSS (ver: 21) statistical package program was used for the calculations.

3. RESULTS

Table 1. Control and change of biochemical parameters studied in chronic arterial disease.

Group **Test**	**CAD** **avg ± SEM** **n=24**	**DM+CAD** **avg ± SEM** **n=12**	**HT+CAD** **avg ± SEM** **n=12**	**Control** **avg ± SEM** **n=12**	**P**
CK (U/L)	67.23 ± 9.12^{b}	135.95 ± 17.91^{a}	73.33 ± 11.86^{b}	40.80 ± 5.72^{b}	0.001
CK-MB (U/L)	22.11 ± 4.31^{b}	45.81 ± 9.99^{a}	19.04 ± 5.38^{b}	14.20 ± 1.56^{b}	0.004
CTnI (NG/ML)	0.60 ± 0.44	6.76 ± 3.78	4.07 ± 4.05	0.023 ± 0.002	0.269

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H-FABP (MG/L)	8.43 ± 1.45^{c}	16.16 ± 0.92m	11.82±0.97^{b}	1.86 ±0.43m	0.050
ALT (U/L)	29.58±5.82^{ab}	39.91 ± 6.08^{a}	22.42±2.91^{b}	18.50±1.45^{b}	0.048
AST (U/L)	31.21 ± 3.96^{b}	89.36 ± 29.36^{a}	31.67±6.10^{b}	23.00±1.34^{b}	0.002
LDH (MG/DL)	298.29±26.81^{a}	292.64±3.04^{a}	318.83±37.42^{a}	160.92±6.87^{b}	0.003
HDL (MG/DL)	50.20 ± 3.69^{a}	36.55 ± 2.90^{b}	51.52± 3.92^{a}	45.63±2.51^{ab}	0.049
LDL (MG/DL)	108.06 ± 6.82	101.75 ± 10.77	90.59±11.44	82.57±5.30	0.143
CREATININE (MG/DL)	0.85 ± 0.18b	1.32 ± 0.25m	1.05± 0.07^{ab}	0.93±0.03^{b}	0.011
GLUCOSE (MMOL/L)	110.04±4.47^{b}	239.36±40.08^{a}	111.92±6.18^{b}	97.42±2.56^{b}	0.001

a,b,c,d The difference between the means shown with different letters in each column is statistically significant

According to these findings, the CK enzyme, which shows changes in muscular destruction, followed a profile similar to the AST level. CK values for the DM+CAD group were calculated to be 3.5 times higher than the controls, and 2 times higher than the other 2 groups (135.95 U/L). This shows how strongly DM is correlated to heart damage (Table 1) ($p \leq 0.001$).

CK-MB, one of the three important isoenzymes of the CK enzyme, is very important as a cardiac marker. A similar table to that of AST-CK can also be found here. The same conclusions reached for the CK analysis can also be used for the interpretation of CK-MB (Table 1). The 14.2 U/L activity observed in the controls reached up to 45.81 U/L in the DM+CAD group ($p\leq 0.004$).
The values for cTn1, which is an important cardiac marker, have also increased significantly in the HT+CAD and DM+CAD groups. As shown in (Table 1), the level was almost zero in the controls, increasing to 6.76 ng/ml in the DM+CAD group. Once again the negative effects of diabetes can be observed ($p\geq 0.269$).

While control group H-FABP level was 1.86, the peak was 11.82 mg/l in the hypertension group and 8.43 mg/l in the CAD group, while it was 16.16 mg/l in the DM+CAD group. This result further emphasizes the negative effect of diabetes on the heart ($p\leq 0.050$).

While the ALT enzyme was measured as 18.50 U/L in the control group, the highest level was found in the DM+CAD group. This shows that diabetes also has negative effects on the liver (Table 1) ($p\leq 0.048$)

AST increases liver or muscle damage and was used as a good diagnostic enzyme to determine heart muscle damage in the past. Here, AST levels were shown to undergo significant changes in CAD cases.The peak value was observed in the DM+CAD group (89.36 U/L) and similar levels were measured in CAD and HT+CAD groups. All three groups had higher AST levels than the controls (Table 1) ($p\leq 0.002$).

The detected HDL-cholesterol levels appear to have decreased, predisposing the patients to heart diseases (Table 1) shows the HDL-cholesterol levels of the other 2 groups, as well as the controls ($p\leq 0.049$).

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LDH levels were increased in the HT+CAD group compared to the control's (318.83 – 160.92 U/L). Similar elevations were found in all 3 groups related to CAD compared to the control ($p \leq 0.003$).

When LDL-cholesterol levels are high, the risk of heart and circulatory diseases increases. Accordingly, (Table 1) has a very good graphical appearance. The lower level in controls (82.57 mg/dl) reached 108.06 mg/dl in the CAD group ($p \geq 0.143$).

Although creatinine, whose clinical significance as a kidney function test is not discussed, did not show much change, the highest value was once again seen in the DM+CAD group (1.32 mg/dl) ($p \leq 0.011$).

As expected, the hunger glucose level was high in the diabetic group and significant changes were detected in the other groups. In Table 1, the value of the DM+CAD group 239.36mmol/L was significantly higher than the others ($p \leq 0.001$).

4. DISCUSSION

The incidence and mortality of cardiovascular disease are increasing worldwide [8] and acute myocardial infarction (AMI) has become an important cause of death [9]. In addition, AMI exhibits a rapid growth trend in young and low-income groups [10]. It is possible to detect these proteins and determine the potential location, severity and course of tissue damage. As such, these are often referred to as biochemical markers, and their correct and quick evaluation is essential to formulate correct treatment and discharge plans. If the rise and fall of these markers in the blood can be detected in time, they can also help discharge or redirect patients who are not actually suffering from tissue damage, or to determine when their recovery is complete. This, in turn, helps reduce hospitalization costs.

That being said, use of such markers accurately to formulate a treatment plan depends on a range of factors. These markers have different release times and rates into the circulatory system, and their elimination rates and paths also vary. Taken both of these into consideration yields the plasma reference levels, which are essential for correct diagnosis and treatment. As such, efforts are focused on identifying tissue-specific proteins that have previously been raised above plasma reference values from biomarker proteins used today. Second, new technologies need to be applied for faster determination of these markers levels to allow for rapid outcomes. In particular, the development of the tests that are applied in the patient room, also known as the point-of-care tests, is attracting a lot of attention, significantly promoting the clinical application of biochemical marker proteins.

Fatty acid-binding proteins (FABPs) are small cytoplasmic proteins that are readily available in large quantities in tissues with active fatty acid metabolisms. Examples of such tissues include the heart and liver. As of this study, nine different types of the FABP families are identified. The most studied is heart-type FABP (H-FABP) largely due to its abundant presence in cardiomyocytes.

Many researchers have studied H-FABP to determine if it can be used as a myocardial damage indicator [5, 11, 12]. Early clinical markers of myocardial infarction include myoglobin (Myo),

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creatine kinase-Mb (CK-MB), cardiac troponin T (cTnT), and cardiac troponin I (cTnI) (13). A new myocardial marker, heart type fatty acid binding protein (H-FABP), has been discovered for the early diagnosis of AMI (14). Under normal physiological conditions, H-FABP is not seen in plasma or tissue fluids and can only be detected in the case of myocardial injury; H-FABP begins to increase 1-3 hours after the onset of acute coronary syndrome. It peaks after 6-8 hours and returns to normal after 12-24 hours. It is one of the earliest markers released into the circulating blood during myocardial injury (15).

All these findings show that, different biomarkers have different utilization windows, and some of this potential can be hidden or lost by the time of initial hospitalization. Many studies have taken this into consideration and compared the potential usability of H-FABP, myoglobin, cTnT, and cTnI for the rapid detection of AMI. The results mostly conclude that H-FABP is an prime candidate as a biomarker for rapid diagnosis and differentiation of clinical events [14]. Table 1 contains the summarized results of 5 similar comparison studies that study how early H-FABP can be used as a marker candidate in hospital-presenting patients with complaints of chest pain suggestive of AMI (Table 1). In each study, the recipient study for accepted blood samples from all patients showed that H-FABP was significantly higher than myoglobin, which demonstrated superior performance.

H-FABP plasma levels usually reach their maximum about 6-8 hours after the first symptoms, and fall back to their normal levels within approximately 1-2 days. Although H-FABP concentration level changes are similar to that of myoglobin (quick to release and clear), its cardiac specificity is approximately 15-20 times greater, so it is a much more effective indicator for myocardial damage. Additionally, the normal serum/plasma levels of H-FABP are significantly lower than that of myoglobin, making it much safer in terms of false-positives.

In some cases, serum H-FABP levels were found to increase within half an hour of ischemic events, making it possible to detect them early. This possibility was tested in numerous studies on bypass graft patients and cardiopulmonary bypass (CPB) pump cases [16,17]. In these studies, it was found that H-FABP has more sensitivity and specificity than troponins and myoglobin. Wider-scope prospective studies were also performed, and in one such study with a cardiac surgery patient count of 1298, various biomarkers like CK-MB, CPB, and H-FABP levels were analyzed. As a result, H-FABP was revealed as an independent marker which could be used to predict death in both the post-op period and beyond [18].

While troponin-I and H-FABP have some a certain degree of variance in terms of their post tissue damage biochemical behaviors, many studies have shown that their plasma levels share similar and often overlapping curves [16,17]. Ischemia can be driven forth as an explanation for this similarity. That being said, neither the reperfusion nor the cytotoxic events caused by neurohormones in plasma are specific to AMI, which also correlates to any troponin-I value beyond the reference. High-sensitivity troponin-I may increase during perioperative myocardial stress as a result of increasing cardiomyocyte apoptosis, stretching of the myocardial wall, or due to proteolysis of the contractile apparatus. None of these cases have myocardial necrosis but high sensitivity troponin-I may increase nonetheless.

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For H-FABP, levels above 26 ng/mL18.2-51.5 ng/mL were found to be a sensitive indicator of ventricular dysfunction, extended hospitalization periods, and mortality rates. One particular study determined a cut-off value of 6.8ng/mL to distinguish myocardial damage in patients undergoing bypass grafting at the pump [19]. The ROC curve-based cutting value was 19.7ng/mL (sensitivity 77%, specificity 75%). The correlation between the various types of biomarkers examined in the present study is slightly different from the studies mentioned above. As a result, it was seen that the largest correlation was found between H-FABP and CK-MB. The correlation between Troponin I and H-FABP is low.

CK-MB is an auxiliary parameter for diagnosis in cases of myocardial injury or infarction. In this study, CK-MB values increased from 14.20 U/L in controls to 45.81 U/L in the DM+CAD group. The most valuable finding of this study is that similar results have been observed in many of the studied parameters related to the heart. The increase in troponin levels is also seen here,where the difference between patient groups and control was statistically significant ($p \leq 0.004$). CK levels also showed a similar profile, resulting in a similar increase in myocardial damage levels ($p \leq 0.001$).

In clinical practices, troponins are now widely adopted as the markers MI detection [20]. However, they are not very well suited to diagnose congestive heart failure and unbalanced angina Pectoris. The same holds true for certain any disorders including ventricular remodeling, or for the cases where extracellular matrix undergoes alterations. Minor myocardial injuries and calcium regulation disorders also challenge the successful use of troponins. Death of cardiac muscle cells was also studied in the literature [21,22]. According to the observations of Setsuda et al., only 48.3% of the CHF patients displayed increased cTnT levels, whereas H-FABP was found to have a success rate of 72.4%. This finding clearly shows that H-FABP was much more sensitive to small cardiomyocyte necrosis compared to cTnT [3].

Another study similar to that of Setsuda et al. was performed with the difference being the tissue damage location. This new study has investigated the differences between sensitivity and accuracy of biomarkers in cases of skeletal muscle injury [23]. The results show that the plasma myoglobin/H-FABP ratio was lower than 1 to 15, proving no correlation between skeletal muscle damage and H-FABP levels (ratio <15) [3]. Therefore, high H-FABP levels only emerge as a result of cardiomyocyte damage.

In that particular study, cTnT levels were found to have raised in CHF and UAP cases, whereas H-FABP levels were found to have stayed the same. This can be explained by the cases being minor MI's, in which H-FABP is rapidly eliminated from the blood but cTnT initially stayed high. This finding is especially profound when we consider the fact that patients displayed no other symptoms that could indicate AMI. This underlies that finding sensitive markers for different tissue damage cases is truly important, and is indicative that H-FABP has a higher sensitivity compared to cTnT for the determination of mild muscle cell damage.

In the presented study, the H-FABP level was measured as 1.86 mg/l in the control group, 11.82 mg/l in the hypertension group, 8.43 mg/l in the CAD group, and 16.16 mg/l in the DM+CAD group ($p \leq 0.050$). Since the difference in the H-FABP mean of the 4 groups examined shows statistical

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importance, this parameter can be suggested as a biomarker in cardiac destructive pathologies. When the H-FABP values were compared proportionally, a 4.5-fold increase was detected in the CAD group compared to the controls. This increase was found to be 6.5 times compared to the HT group and 8.7 times to the DM+CAD group. These elevations seem to be important as a descriptor of a heart problem that will be shaped by different reasons in the patient and will help guide the clinical diagnosis.

Current clinical norm is that ALT and AST tests are the primary methods of determining and monitoring the course of hepatocellular injuries and various other types liver diseases [24]. Current liver function tests are sufficiently specific for liver disease despite their lack of sensitivity. While it's true that ALT analysis has become rapid, cheap and easily accessible thanks to wide range of clinical analyzers, it still has the issue that ALT is a large molecule (96 kD) and it only starts to increase in circulation after severe cell damage or death. Various cytoplasmic proteins share the same problem, but owing to the fact that hepatocytes lack an interstitial barrier due to their large endothelial clefts and are in close contact with vasculature, protein with smaller molecular size diffuse in plasma faster compared to their large counterparts, as in damaged cardiomyocytes, and therefore increase beyond their normal values earlier in serum compared to their larger counterparts. All things considered, new specific and sensitive liver injury markers are generally needed for acute hepatocellular injury diagnosis and treatment [24].

α-GST has been suggested as one such marker for hepatocellular damage due to its strong sensitivity and specificity. Found in the liver, kidneys, and intestines, this cytoplasmic 26 kD protein is quick to be released to the circulation from even slightly-damaged liver cells. α-GST also has a relatively shorter plasma presence [25,26]. That being said it's not specificity is still not as sharp as that of H-FABP, as this has been shown in some studies [20] . Still, α-GST was shown to be a great biomarker in post liver-transplant rejection cases [2].

In the present study, when the ALT level was examined, the highest activity was found in the DM + CAD group. As can be seen in Table 1, the intergroup significance was determined at the level of $p<0.048$. While a value close to that of the controls was determined in the HT-CAD group, higher values were calculated in the CAD group.

Creatinine levels are important for kidney function. In this study, creatinine level was highest in the DM+CAD group. The nephropathic effect of diabetes is underlined by this finding. In the statistical interpretation of the mean between the groups, importance was found at the level of $p\leq0.011$ (Table 1).

Considering all of these, H-FABP stands above others as an early cardiac tissue damage marker. Smaller proteins are released to the circulation faster than their larger counterparts in case of cell damage [4], and H-FABP is lighter compared to many other markers. Another factor influencing the rate of increase of a marker in circulation is its amount in the cells, the more a tissue contains that particular marker, the faster it will rise beyond its reference serum value. Therefore, the best marker for the detection of injuries on a given tissue has to be specific to that particular tissue [27,28]. It should also be protein with a relatively smaller molecular size, as this is usually related to early

release. Similarly, the marker should be readily available on that particular tissue, while simultaneously having a low plasma presence in healthy individuals. H-FABP meets all these criteria for cardiac muscle damage.

Troponins are also good candidates due to their good cardiac specificity, however they are still late markers compared to H-FABP [29]. This delay is due to the fact that they are broken down from tropomyosin, which takes time. Another shortcoming of troponins is their extended plasma presence, which may last days.

LDH enzyme activity showed a lot of variation in this study. An LDH level of 160.92 U/L was determined in the control group, which was 298.29, 292.64,and 318.83 U/L in the CAD, DM+CAD, and HT+CAD groups respectively($p \leq 0.003$).

H-FABP and myoglobin are similar in many regards regarding cardiac tissue damage. They both increase beyond their reference values rapidly, and they both are eliminated from the circulation quickly through the kidney path. This makes both elements good candidates for early markers, from the onset of symptoms to approximately 1-2 days. Their plasma levels reach the peak within 6 - 12 hours of MI, and return to normal within 1 day (36 hours if no thrombolytics are used). However it's still possible to consider H-FABP as a better choice compared to myoglobin as the preferred early cardiac marker [20]. The level difference of H-FABP concentration in healthy and CAD individuals [30] and the high sensitivity of H-FABP to minor myocardial damages, will enable it to be used in diagnosis - even in patients with chronic heart failure or unbalanced angina Pectoris [31,32].

FABP seems to be a bright candidate for future rapid tissue damage diagnosis parameters. While some members of the family are not tissue-specific (H and L-FABP), they are still reported as the most sensitive early diagnosis markers for myocardial, bone, kidney, brain, liver, and intestine damage. Other members of FABP family are tissue specific, where L-FABP shows liver damage, while I-FABP shows intestine and B-FABP shows brain damage.

New rapid testing systems that implement specific monoclonal antibodies and antigens will enable FABP to become a very rapid diagnosis tool in clinics, especially in conjunction with other clinical findings. This can lead to reduced mortality and morbidity.

In the presented study, the changes in biochemical parameters due to combinations of CAD with different diseases and conditions (Hypertension and Diabetes) were examined. The most severe changes in the studied parameters were observed in individuals with diabetes. The increase in H-FABP was mostly shaped in the Diabetic group, and the differences in the parameters between the groups were important for all markers, except for cTnI and LDL. These are important results for cardiological studies.

As a result: clinically, CAD and MI are diagnosed with biochemical measurements such as troponin, CK-MB, LDH, AST, CK, and early treatment plans were shaped according to such findings. The results of this study, however, show that H-FABP levels in different groups such as CAD, DM+CAD, and HT+CAD had changed significantly compared to the control. This means that H-FABP can be

used as a biochemical cardiomarker, and should be taken into account from the first minutes of damage. Due to its early occurrence, specificity and sensitivity compared to other cardiac markers as stated by the literature and as shown here, it can be included in routine biochemistry tests. We hope that our findings will contribute to H-FABP being considered in clinical use.

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APPENDIX

T.C.
VAN YÜZÜNCÜ YIL ÜNİVERSİTESİ
GİRİŞİMSEL OLMAYAN
KLİNİK ARAŞTIRMALAR ETİK KURULU
KARAR FORMU

	Prof. Dr. Nihat MERT sorumluluğunda yapılması tasarlanan ve yukarıda başvuru bilgileri verilen "Kronik Arter Hastalıklarında Kalp Tipi Yağ Asidi Bağlayıcı Protein (H-FABP) ve Bazı Biyokimyasal Parametrelerin Değişimi" isimli bilimsel araştırma başvuru dosyası ve ilgili belgeler araştırmanın gerekçe, amaç, yaklaşım ve yöntemleri dikkate alınarak incelenmiştir. Araştırmacıların Van Yüzüncü Yıl Üniversitesi Girişimsel Olmayan Klinik Araştırmalar Etik Kurulunun Çalışma Esasları Hakkında Yönergesinde belirtilen hususları yerine getirdikleri belirlenmiş olup, çalışmaları ile ilgili tüm sorumluluk araştırmacılara ait olmak üzere, söz konusu çalışmanın gerçekleştirilmesinde sakınca bulunmadığına, toplantıya katılan Etik Kurul üye tam sayısının salt çoğunluğu/oy birliği ile karar verilmiştir.
GİRİŞİMSEL OLMAYAN KLİNİK ARAŞTIRMALAR ETİK KURULU	
ETİK KURULUN ÇALIŞMA ESASI	Klinik Araştırmalar Hakkında Yönetmelik, İyi Klinik Uygulamaları Kılavuzu
BAŞKANIN UNVANI / ADI / SOYADI:	**Prof. Dr. Yasin TÜLÜCE**

Unvanı/Adı/Soyadı	Uzmanlık Alanı	Kurumu	Cinsiyet		Araştırma ile ilişki		Katılım *		İmza
Prof. Dr. Yasin TÜLÜCE	Tıbbi Biyoloji	Van Yüzüncü Yıl Üniversitesi Tıp Fakültesi	E ☒	K ☐	E ☐	H ☒	E ☒	H ☐	
Prof. Dr. Sıddık KESKİN	İstatistik Uzmanı	Van Yüzüncü Yıl Üniversitesi Tıp Fakültesi	E ☒	K ☐	E ☐	H ☒	E ☒	H ☐	
Prof. Dr. Özgür KEMİK	Genel Cerrahi	Van Yüzüncü Yıl Üniversitesi Tıp Fakültesi	E ☒	K ☐	E ☐	H ☒	E ☐	H ☒	
Doç. Dr. Serap GÜNEŞ BİLGİLİ	Dermatoloji	Van Yüzüncü Yıl Üniversitesi Tıp Fakültesi	E ☐	K ☒	E ☐	H ☒	E ☒	H ☐	
Doç. Dr. Mahmut SÜNNETÇİOĞLU	Klinik Bakteriyoloji ve Enfeksiyon	Van Yüzüncü Yıl Üniversitesi Tıp Fakültesi	E ☒	K ☐	E ☐	H ☒	E ☐	H ☒	
Dr. Öğr. Üyesi Mahammed BATUR	Göz Hastalıkları	Van Yüzüncü Yıl Üniversitesi Tıp Fakültesi	E ☒	K ☐	E ☐	H ☒	E ☒	H ☐	
Dr. Öğr. Üyesi Emine TÜRKMENOĞLU	Deontoloji	Van Yüzüncü Yıl Üniversitesi Veteriner Fakültesi	E ☐	K ☒	E ☐	H ☒	E ☒	H ☐	
Dr. Öğr. Üyesi Orxe ALLAHVERDİYEV	Tıbbi Farmakoloji	Van Yüzüncü Yıl Üniversitesi Eczacılık Fakültesi	E ☒	K ☐	E ☐	H ☒	E ☒	H ☐	
Dr. Öğr. Üyesi Zehra KAYA	Tıbbi Biyoloji	Van Yüzüncü Yıl Üniversitesi Tıp Fakültesi	E ☐	K ☒	E ☐	H ☒	E ☐	H ☒	
Dr. Öğr. Üyesi Sermin ALGÜL	Fizyoloji	Van Yüzüncü Yıl Üniversitesi Tıp Fakültesi	E ☐	K ☒	E ☐	H ☒	E ☒	H ☐	
Dr. Öğr. Üyesi Özgür GENÇ ŞEN	Endodonti	Van Yüzüncü Yıl Üniversitesi Diş Hekimliği Fakültesi	E ☐	K ☒	E ☐	H ☒	E ☒	H ☐	
Nazlı AKTAŞ	Avukat	Van Yüzüncü Yıl Üniversitesi Hukuk Müşavirliği	E ☐	K ☒	E ☐	H ☒	E ☒	H ☐	
Lütfu POLAT	Eczacı	Van Polat ECZANESİ	E ☒	K ☐	E ☐	H ☒	E ☒	H ☐	
Özge Burak DEĞER	Sağlık Meslegi Mensubu Olmayan Üye	Van Sanayiciler ve İş Kadınları Derneği	E ☐	K ☒	E ☐	H ☒	E ☒	H ☐	
Adnan SELÇUK	Sağlık Meslegi Mensubu Olmayan Üye	Van İş Geliştirme Merkezi	E ☒	K ☐	E ☐	H ☒	E ☐	H ☒	

Sayfa 2

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