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NT-proBNP as an Additional Marker of Significant Coronary Atherosclerotic Lesions

Abstract. Coronary artery disease (CAD) is the leading cause for morbidity and mortality both in Ukraine and in the world, so the relevance of this problem for the society is undeniable. The priority is still to study the factors that affect both more severe CAD in patients with chronic coronary syndrome and after myocardial revascularization.

The aim. To investigate the patterns of correlation between blood level of N-terminal pro-B-type natriuretic peptide (NT-proBNP) and CAD severity, especially in patients with multivessel lesion, to confirm its status of a useful additional marker for assessing the condition of cardiovascular system and one of the instruments to affect the tactics of treatment.

Materials and methods. The study is based on data obtained from a prospective analysis of 40 patients at the age of 51 to 82 years old from January to December 2019, whose complaints could indicate the CAD. All the patients underwent a comprehensive clinical and laboratory examination (complete blood count, biochemical blood test). The main instrumental examination method was coronary angiography; the patients were divided into 2 groups on the basis of the examination results. The quantitative degree of lesion was assessed using the SYNTAX Score for each patient.

Results. The groups were comparable in terms of age, sex, and comorbidities. The groups differed significantly in terms of body mass index. Moreover, the groups differed in the level of the following biochemical markers: NT-proBNP ($p=0.0001$), cholesterol ($p=0.02$), low-density lipoproteins ($p=0.009$), creatinine ($p=0.02$), glomerular filtration rate ($p=0.08$). A significant correlation was found between the NT-proBNP level and the degree of CAD $\rho=0.718$ ($p=0.0001$).

Conclusion. NT-proBNP significantly correlates with the SYNTAX Score and is the highest in the group of patients with multivessel coronary disease. This indicator requires further study as an additional marker for assessing the state of the cardiovascular system and can influence the choice of treatment.

Keywords: coronary artery disease, homocysteine, body mass index, NT-proBNP, multivessel disease, SYNTAX Score.

Introduction. Coronary artery disease (CAD) is the leading cause for morbidity and mortality both in Ukraine and in the world [5], so the relevance of this problem for the society is undeniable. Since the Framingham study, enormous amount of information has been accumulated on calculating risks, potential hazards of certain factors and diseases, such as age, gender, genetic factors, smoking, high blood pressure and type 2 diabetes mellitus. The multifactorial nature of atherosclerosis, as the root cause of CAD, is beyond doubt. However, the decision-making in each case requires a comprehensive assessment of not only coronary arteries, but also biochemical markers and patient's comorbidity. The SYNTAX Score is often used to quantify the degree of coronary vessels damage [12]. So, for example, in patients with a higher score on this scale (more than 22) and diabetes mellitus, coronary artery bypass grafting (CABG) is more preferable than percutaneous coronary intervention (PCI) as a method of revascularization.

Factors of CAD progression have not been fully studied for patients receiving drug treatment after PCI or CABG.

The role of markers such as troponin, homocysteine and C-reactive protein (CRP) in the disease progression has already been proven [8, 14].

Recently, there has been increasing evidence that quantification of the presence and extent of cardiac haemodynamic stress and heart failure is performed with the use of N-terminal pro-B-type natriuretic peptide (NT-proBNP).

The level of this marker can help in the choice of treatment (PCI or CABG) as a method of revascularization in patients with the ostial left main lesion and/or three-vessel coronary artery disease [7].

Since 1981, when de Bold et al. described a substance causing natriuresis in rats, a family of peptides with similar properties has been identified [4].

Testing for NT-proBNP, one of the representatives of this family, is currently mandatory for patients with acute and chronic heart failure for more accurate verification of the diagnosis and prognosis. According to European recommendations, the upper limit of NT-proBNP is <125 pg/ml in non-acute situation and 300 pg/ml in the acute situation [9]. The trigger for release is stretching of myocardial fibers, while the highest concentration is observed at a volume overload of heart chambers. The level also increases in acute coronary syndrome, myocardial infarction, myocarditis, myocardial hypertrophy. At the same time, increase in NT-proBNP concentration provides additional information about adverse outcomes such as death, myocardial infarction, and stroke [2]. In addition, increased concentrations of this marker may be useful in making decisions about the choice of effective revascularizations methods (CABG or PCI) [11].

On the other hand, PCI is a preferable option for patients without heart failure and, accordingly, with the normal or slightly elevated NT-proBNP concentrations. Recently, the increase in NT-proBNP levels has been studied as a severity factor of CAD in patients with chronic coronary syndrome and is determined as a prognostically unfavourable factor after myocardial revascularization [7].

The aim of this study was to investigate the patterns of correlation between blood level of NT-proBNP and CAD severity, especially in patients with multivessel lesion, to confirm its status of a useful additional marker for assessing the condition of cardiovascular system and one of the instruments to affect the tactics of treatment.

Materials and methods. The study was performed as a prospective observation at the Ukrainian Children's Cardiac Center, Clinic for Adults (Kyiv) during 2019. The data were obtained from 40 patients eligible for coronary angiography. Based on the results of coronary angiography, the patients were divided into 2 groups. The control group (group I) consisted of 10 patients without CAD. The study group (group II) included 30 patients with varying degrees of CAD (in particular, nonstenotic lesion, single-vessel and multivessel CAD).

Subsequently, group II was divided into 2 subgroups: subgroup IIA consisted of patients with non-stenotic atherosclerosis and single-vessel disease (n=15), subgroup IIB included patients with multivessel CAD (n=15).

The groups were comparable in terms of age, sex, and comorbidities, but the body mass index (BMI) was significantly different. In the first group of patients, there was a higher BMI, although the average value and median of both groups corresponded to the first stage of obesity. The patients' distribution within the groups was as follows. Group I: 1 patient (10%) with normal BMI, 1 (10%) overweight, 3 (30%) with stage 1 obesity, 5 (50%) with stage 2 obesity. Group II: 3 patients (10%) with normal BMI, 10 (33.3%) overweight, 12 (40%) with stage 1 obesity, 4 (13.3%) with stage 2 obesity, 1 (3.3%) with stage 3 obesity. The data indicate a higher percentage of stage 2 obesity (50%) in patients without vessels damage. In group II, the majority of patients were overweight (33.3%) and with stage 1 obesity (40%).

According to the criteria of the New York Heart Association, group II had the following distribution by functional classes (FC): 8 (26.7%) patients had FC I, 18 (60%) had FC II, and 14 (13.3%) had FC III heart failure. Upon further division into subgroups (patients with non-stenotic atherosclerosis and single-vessel disease (IIA) and patients with multivessel disease (IIB), there was no significant difference according to the FC by the New York Heart Association (p=0.53). The data are shown in Table 1.

In group I, 5 (50%) patients did not receive lipid-lowering therapy, 3 (30%) patients received atorvastatin 20 mg once daily, and 2 (20%) patients received rosuvastatin 10 mg once daily. Seven (23.3%) patients from group II did not receive statin treatment, 1 (3.3%) patient took simvastatin 40 mg, 11 (36.7%) patients took atorvastatin 40 mg once daily, 11 (36.7%) of patients took rosuvastatin 20 mg.

The exclusion criteria were atrial fibrillation/flutter, severe insufficiency or stenosis of valves, all types of cardiomyopathies, myocarditis of different origin, acute inflammatory diseases, hormonal therapy, and pregnancy.

Patients with thyroid diseases were in a state of euthyroidism which was confirmed by the level of blood hormones according to laboratory studies. This investigation was performed at the prehospital stage. Patients did not receive hormone replacement therapy at the time of hospitalization.

The study was conducted according to the principles of the Declaration of Helsinki. The written informed consent was approved by the review board of the Ukrainian Children's Cardiac Center and later signed by all the participants.

All the patients underwent clinical examination and laboratory testing. Venous blood samples were taken from each subject and were subsequently centrifuged prior to testing. Blood tests (white blood cells, neutrophils, lymphocytes, monocytes, red blood cells, platelets and hemoglobin) and hematocrit measurements were performed using ABX Pentra 60 C+ hematology analyzer (HORIBA ABX, Montpellier, France). For complete blood count and erythrocyte sedimentation rate determination, venous blood

Table 1

Baseline characteristics of the study population

Characteristics	Group I (n= 10)	Group II (n= 30)	p value*
Age (years), mean±SD	61.9±2.43	66±1.45	p=0.2
Males, n (%)	4 (40%)	19 (63.3%)	p=0.2
BMI (kg/m^2), mean±SD	33.78±1.3	30.65±0.98	p=0.03
Previous myocardial infarction, n (%)	0	14 (46.7%)	p=0.001
Stroke or TIA history, n (%)	1 (10%)	5 (16.7%)	p=0.6
Smoking, n (%)	6 (60%)	10 (33.3%)	p=0.1
Family history of CAD, n (%)	6 (60%)	15 (50%)	p=0.6
Hypertension, n (%)	10 (100%)	30 (100%)	p=1.0
Diabetes mellitus, n (%)	2 (20%)	6 (20%)	p=1.0
Atrial fibrillation, n (%)	2 (20%)	3 (10%)	p=0.4
COPD, n (%)	2 (20%)	5 (16.7%)	p=0.8
Thyroid disease, n (%)	2 (20%)	9 (30%)	p=0.5
β-blockers, n (%)	6 (60%)	18 (60%)	p=1.0
Calcium antagonists, n (%)	3 (30%)	7 (23.3%)	p=0.7
ACE inhibitors / sartans, n (%)	4 (40%)	23 (76.7%)	p=0.03
Acetylsalicylic acid, n (%)	5 (50%)	20 (66.7%)	p=0.4
Clopidogrel, n (%)	1 (10%)	16 (53.3%)	p=0.02

Values are presented as mean ± SD or number (%).
*p<0.05 is statistically significant.
ACE, angiotensin converting enzyme; COPD, *chronic obstructive pulmonary disease;* SD, standard deviation; TIA, *transient ischemic attack.*

samples were collected from each patient in blood tubes containing ethylenediaminetetraacetic acid or citrate. Biochemical analyses (total protein, CRP, fasting blood glucose, alanine aminotransaminase (ALT), aspartate aminotransferase (AST), blood urea, creatinine, total bilirubin, calcium, potassium, sodium, fasting triglyceride, total cholesterol, low-density lipoprotein cholesterol (LDL-C), and high-density lipoprotein cholesterol (HDL-C)) were performed using AU 480 chemistry analyzer (Beckman Coulter, Brea, CA, United States). All the tests were performed according to the manufacturers' instructions. The level of NT-proBNP was analyzed with enzyme-linked fluorescence assay by automatic miniVIDAS® (bioMerieux, Craponne, France). Samples for coagulation tests which included prothrombin index and international normalization ratio were investigated using Thrombotimer 4 (Behnk Elektronik, Norderstedt, Germany).

Coronary angiography was performed on Artis Zee biplane system (SIEMENS, Germany).

Statistical analysis. All statistical analyses were performed using the licensed computer program STATISTICA v.6.1 (Statsoft Inc., USA, serial number AGAR909

E415822FA). The obtained data were analyzed using the Shapiro-Wilk test. Continuous variables were non-normally distributed and are presented as a median and 25-75% interquartile range. Categorical variables are presented as number and percentage. Differences in participant characteristics were compared with the Pearson χ^2 test for categorical variables, and with the analysis of variance or the Mann-Whitney U test for continuous variables. Spearman and Pearson's correlation coefficient was used to estimate the relationships. Statistically significant results were considered if p<0.05 [1].

Results and discussion. The analysis of homocysteine showed that 7 (70%) patients from group I and 22 (73.3%) from group II had normal levels. Elevated rates were found in 3 (30%) patients from group I and 8 (26.7%) patients from group II. These data indicate that the biochemical parameter is doubtful and may not be a risk factor for the development of CAD. After an assessment, it is necessary to take into account conditions in which this marker may be increased: thrombosis, endothelial dysfunction, genetic defects of enzymes involved in homocysteine metabolism, renal failure, hypothyroidism, B12 deficiency anemia.

Currently, there is a widespread theory that the increase of blood homocysteine is associated with higher cardiovascular risk, regardless of other factors for atherosclerotic lesions [3]. From a pathophysiological point of view, hyperhomocysteinemia is associated with increased thrombogenicity, oxidative stress due to activation of redox inflammatory pathways and impaired endothelial function which is the root cause of atherogenesis.

In contrast, other clinical trials, such as NORVIT and HOPE-2 [6], did not show a reduction in cardiovascular events with decreased homocysteine levels in patients with atherosclerotic lesions. Therefore, the use of homocysteine as a reliable marker of cardiovascular events remains debatable.

Comparative characteristics of other biochemical markers in patients of groups I and II are shown in Table 2.

There were no significant differences between the following parameters: fasting glucose, total bilirubin, ALT, AST, CRP, triglycerides, HDL-C, and urea.

There were significant difference between the groups in terms of NT-proBNP, cholesterol, LDL-C, creatinine, creatinine clearance (calculated with the modification of diet in renal disease, MDRD).

The median NT-proBNP in group I was 65.45 pg/ml (Q1-Q3: 39.35–97.65 pg/ml) which is significantly lower than that in group II where it reached 539.2 pg/ml (Q1-Q3: 167.48–1260.75 pg/ml), p=0.0001. Median cholesterol level in patients without vascular lesions was 5.3 mmol/L (Q1-Q3: 4.35–5.9 mmol/L), and in those with atherosclerotic lesions it was lower: 3.75 mmol/L (Q1-Q3: 3.4–4.7 mmol/L), p=0.02. Similar data were obtained for LDL-C. In group I, the result was higher: median 3.61 mmol/L

Table 2

Biochemical parameters

Parameter	Group I (n = 10)	Group II (n = 30)	p value*
Homocysteine, µmol/L	13.10 (10.95–20.55)	14.6 (12.83–18.13)	p=0.66
Total protein, g/L	77.10 (68.65–79.15)	72.5 (69.78–76.68)	p=0.37
Glucose (fasting), mmol/L	6.20 (5.20–6.93)	5.95 (5.7–6.9)	p=0.76
Total bilirubin, µmol/L	14.3 (11.5–22.6)	14.65 (12.25–19.98)	p=0.66
ALT, U/L	34.25 (17.28–40.18)	28.4 (17.7–39.35)	p=0.64
AST, U/L	20.6 (15.25–31.85)	23.1 (18.03–30.88)	p=0.6
NT-proBNP, pg/mL	65.45 (39.35–97.65)	539.2 (167.48–1260.75)	p=0.0001
CRP, mg/L	3.48 (2.38–4.98)	3.25 (1.78–9.65)	p=0.66
Total cholesterol, mmol/L	5.3 (4.35–5.9)	3.75 (3.4–4.7)	p=0.02
Triglyceride, mmol/L	1.57 (1.27–2.16)	1.14 (0.94–1.82)	p=0.11
HDL, mmol/L	1.1 (0.92–1.37)	1.04 (0.94–1.82)	p=0.44
LDL, mmol/L	3.61 (2.98–3.79)	2.2 (1.93–2.93)	p=0.009
Urea, mg/dL	5.85 (4.9–7.23)	5.9 (5.48–6.93)	p=0.93
Creatinine, µmol/L	86.6 (79.95–99.52)	102.6 (88.88–122.35)	p=0.02
CrCl (mL/min/1.73 m^2)**	69.5 (62.5–77.75)	62 (50.75–70.25)	p=0.08

Values are presented as median (25th percentile – 75th percentile).
*p<0.05 is statistically significant.
**Calculated with the modification of diet in renal disease.
CrCl, creatinine clearance.

(Q1-Q3: 2.98–3.79 mmol/L) versus 2.2 mmol/L in group II (Q1-Q3: 1.93–2.93 mmol/L), p=0.009.

The data may have some incompatibility with the general theory of atherogenesis, but endothelial dysfunction [3] and oxidative stress are more likely to play a leading role in the development of systemic atherosclerotic lesions.

The median creatinine level in group I was 86.6 µmol/L (Q1-Q3: 79.95–99.52 µmol/L), median creatinine clearance according to the MDRD was 69.5 ml/min/1.73 m^2 (Q1-Q3: 62.5–77.75 mL/min/1.73 m^2). In group II, the rate was significantly higher and amounted to median 102.6 µmol/L (Q1-Q3: 88.88–122.35 µmol/L), the creatinine clearance (MDRD) was reduced — 62 ml/min/1.73 m^2 (Q1-Q3: 50.75–70.25 ml/min/1.73 m^2), p=0.08. The data

has confirmed the association of cardiovascular diseases and adverse consequences in patients with the impaired renal function [10, 13].

To identify the correlations between biochemical parameters and the progression of atherosclerosis, group II was divided into two subgroups. IIA subgroup consisted of patients with non-stenotic atherosclerosis and single-vessel lesions (15 patients), IIB subgroup included those with multivessel lesions of the coronary arteries (15 patients). The data are shown in Table 3.

The results of cholesterol metabolism in patients from subgroups IIA and IIB were similar. Median cholesterol in patients with multivessel lesions was 4.2 mmol/L (Q1-Q3: 3.5–4.7 mmol/L), and low-density lipoprotein was 2.52 mmol/L (Q1-Q3: 1.94–3.01 mmol/L). It should be noted

Table 3

Comparison of biochemical markers

Parameter	Group I (n= 10)	Subgroup IIA (n= 15)	p value*	Subgroup IIB (n= 15)	p value**
NT-proBNP, pg/mL	65.45 (39.35–97.65)	232.0 (80.6–780.0)	p=0.006	713.0 (495.0–1420.0)	p<0.0001
Total cholesterol, mmol/L	5.3 (4.35–5.9)	3.7 (3.4–4.7)	p=0.03	4.2 (3.5–4.7)	p=0.05
LDL-C, mmol/L	3.61 (2.98–3.79)	2.05 (1.89–2.79)	p=0.01	2.52 (1.94–3.01)	p=0.05
Creatinine, µmol/L	86.6 (79.95–99.52)	98.0 (88.2–106.7)	p=0.08	108.1 (90.8–123.1)	p=0.02
CrCl (mL/min/1.73 m^2)***	69.5 (62.5–77.75)	66.0 (53.4–76.0)	p=0.4	55.0 (49.0–70.0)	p=0.04

Values are presented as a median (25th percentile – 75th percentile).
*Results comparison between group I and subgroup IIA; the difference in parameters is statistically significant (p<0.05).
**Results comparison between group I and subgroup IIB; the difference in parameters is statistically significant (p<0.05).
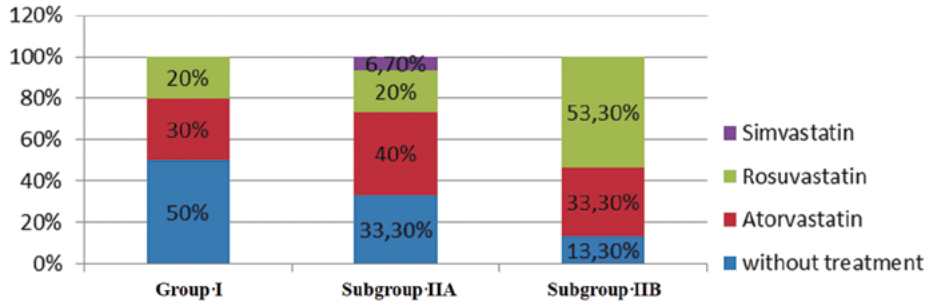***Calculated with the modification of diet in renal disease (MDRD).

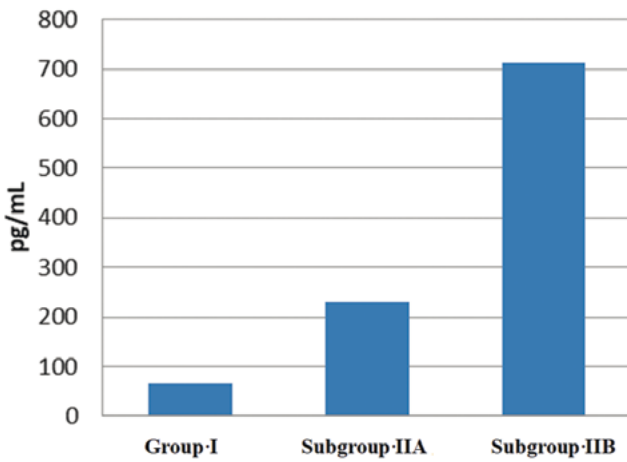**Fig. 1.** *Statin therapy in different groups of patients*

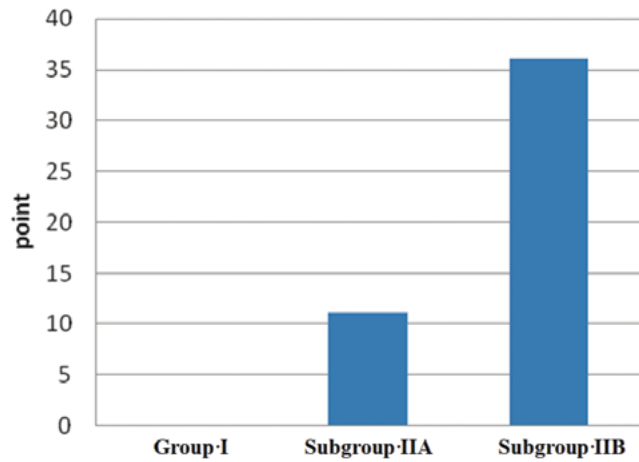**Fig. 2.** *Level of NT-proBNP*

Fig. 3. *SYNTAX Score*

that lipid-lowering drugs primarily affected the parameters of cholesterol metabolism (Fig. 1). The results indicated ineffective dosing of drugs to avoid or slow down atherosclerosis.

Creatinine levels were the lowest in group I. The average value was 88.66 μmol/L (Q1-Q3: 79.95–99.52 μmol/L), creatinine clearance with MDRD was reduced to 69.5 mL/min/1.73 m^2 (Q1-Q3: 62.5-77.75 mL/min/1.73 m^2), which corresponds to stage 2 of chronic kidney disease (CKD). With the progression of CAD, the level of nitrogen wastes increases. In subgroup IIA, creatinine was 98.0 μmol/L (Q1-Q3: 88.2–106.7 μmol/L) and creatinine clearance with MDRD was 66.0 mL/min/1.73 m^2 (Q1-Q3: 53.4-76.0 mL/min/1.73 m^2) which corresponds to mild kidney damage. Patients in subgroup IIB had the highest creatinine value of 108.1 μmol/L (Q1-Q3: 90.8-123.1 μmol/L) and the lowest creatinine clearance with MDRD 55.0 mL/min/1.73 m^2 (Q1-Q3: 49.0-70.0 mL/min/1.73 m^2), which complies to stage 3 CKD and indicates cardiorenal syndrome.

Subgroups IIA and IIB differed in terms of anatomical SYNTAX Score. Subgroup IIA had a low risk according to SYNTAX Score: median 10 points (Q1-Q3: 5-16 points). Subgroup IIB showed median 35 points (Q1-Q3: 26-43.5 points) which evidences a high risk.

This result showed a significant correlation between the level of NT-proBNP and the degree of CAD (according to SYNTAX Score) ρ=0.718, p<0.0001. The data are shown in Fig. 2 and 3.

Conclusion

1. NT-proBNP significantly correlated with anatomical SYNTAX Score and was the highest in the group of multivessel CAD. This study shows NT-proBNP as an independent marker of more significant coronary vessels lesions. It can be used as a predictor of not only more significant vascular lesion but also assessment for treatment after PCI and CABG. Further research is feasible.

2. The importance of homocysteine has been revealed as an independent modified factor in the development of cardiovascular disease. Increased homocysteine can lead to the progression of atherothrombotic, cerebral, cardiac, or peripheral vascular disorders, as well as neurodegenerative processes. However, our investigation shows that the level of homocysteine was the same in patients of the control and study groups, which confirms the controversial data on its effect on the processes of atherosclerosis occurrence and progression.

3. Total cholesterol and LDL-C were highest in patients of control group, as well as the number of patients

not treated with lipid-lowering drugs. BMI also did not affect the degree of damage to the coronary vessels. This may indicate that oxidative stress and endothelial dysfunction are paramount in the process of atherogenesis.

Conflicts of interest.
Authors declare no conflict of interests.

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NT-proBNP як додатковий маркер більш значущого атеросклеротичного ураження вінцевих судин

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Резюме. Ішемічна хвороба серця займає найвищі позиції щодо захворюваності та смертності в Україні і в світі, тому актуальність цієї проблеми беззаперечна як для ранньої діагностики, так і для ефективного лікуван-

ня. Пріоритетним залишається вивчення факторів, які впливають на більш тяжке ураження вінцевих артерій у пацієнтів з хронічним коронарним синдромом, а також після реваскуляризації міокарда.

Мета дослідження. Проаналізувати кореляцію рівня NT-proBNP та інших біохімічних маркерів у крові з вираженістю ішемічної хвороби серця, зокрема у пацієнтів із багатосудинним ураженням, щоб підтвердити його необхідність як додатковий маркер для оцінювання стану серцево-судинної системи та одного з інструментів, що впливають на тактику лікування.

Матеріали та методи. В основу дослідження покладено відомості, отримані під час проспективного аналізу даних 40 пацієнтів віком від 51 до 82 років у період із січня по грудень 2019 року, характер скарг яких міг свідчити про наявність ішемічної хвороби серця. Всім пацієнтам було проведено комплексне клініко-лабораторне дослідження (загальний аналіз крові, біохімічний аналіз крові). Основний інструментальний метод дослідження – коронаровентрикулографія, за результатами якої пацієнти були розділені на 2 групи. Для кожного пацієнта визначено кількісний ступінь ураження за допомогою шкали SYNTAX Score.

Результати та їх обговорення. Групи були зіставні за віком, статтю та наявними супутніми захворюваннями. За індексом маси тіла групи достовірно відрізнялися. Також групи відрізнялися за рівнем таких біохімічних маркерів: NT-proBNP (р = 0,0001), холестерин (р = 0,02), ліпопротеїди низької щільності (р = 0,009), креатинін (р = 0,02), швидкість клубочкової фільтрації (р = 0,08). Виявлено значущу кореляцію між рівнем NT-proBNP і ступенем ураження вінцевих судин ρ = 0,718 (р < 0,0001).

Висновок. NT-proBNP достовірно корелює з кількістю балів за шкалою SYNTAX Score і був найвищим у групі пацієнтів з багатосудинним ураженням вінцевих судин. Цей показник вимагає подальшого вивчення як додаткового маркера оцінювання стану серцево-судинної системи і в подальшому може вплинути на вибір тактики лікування.

Ключові слова: ішемічна хвороба серця, гомоцистеїн, індекс маси тіла, NT-proBNP, багатосудинне ураження, SYNTAX Score.

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