MINISTRY OF EDUCATION AND SCIENCE OF UKRAINE
SUMY STATE UNIVERSITY
ACADEMIC AND RESEARCH MEDICAL INSTITUTE

Eastern Ukrainian Medical Journal

116, Kharkivska st., Sumy 40007, Ukraine eumj.med.sumdu.edu.ua
e-mail: eumj@med.sumdu.edu.ua ISSN: 2663-5909 (print)/2664-4231 (online)

How to cite / Як цитувати статтю: Melnychuk I. Gut microbiota metabolites connections with echocardiography atrial fibrillation characteristics in patients with coronary artery disease. *East Ukr Med J*. 2024;12(1):137-147

DOI: https://doi.org/10.21272/eumj.2024;12(1):137-147

ABSTRACT

Iryna Melnychuk
https://orcid.org/0000-0002-0659-1476
Internal Medicine Department No. 4, Bogomolets National Medical University, Kyiv, Ukraine

GUT MICROBIOTA METABOLITES CONNECTIONS WITH ECHOCARDIOGRAPHY ATRIAL FIBRILLATION CHARACTERISTICS IN PATIENTS WITH CORONARY ARTERY DISEASE

The aim: To reveal the connections between gut microbiota metabolites and echocardiography (TTE) atrial fibrillation (AF) characteristics in patients with coronary artery disease (CAD).

Materials and methods: 300 patients were divided into 3 groups: the first group(CAD) – 149 patients with CAD but without arrhythmias, the second group (CAD+AF) – 124 patients with CAD and AF paroxysm, and the control group – 27 patients without CAD and arrhythmias. TTE was done by ALOKA SSD-5000. The level of trimethylamine (TMA), trimethylamine-N-oxide (TMAO) of plasma, and fecal short-chain fatty acids (SCFA) levels were determined by gas chromatography with mass electron detection.

Results: Isocaproic and isobutyric fecal acids appear in group II in comparison with CG. In group II, patients' rise of TMA (16.13%), TMAO (57.54%) levels, and decreased ratio (26.16%) in comparison with group I was revealed, $P<0.05$. In group II patients rise of valeric (1128.43%) and depletion of butyric (78.75%), isovaleric (43.71%), caprylic (99.21%) acids, middle chain fatty acids (95.54%), and the total amount of fecal SCFA (17.09%) in comparison with group I was found, $P<0.05$. In group II, patients' rise in left atrium diameter (10.03%), left atrium volume (15.40%), and left atrium volume index (11.48%) in comparison with group I was revealed, $P<0.05$. The largest amount of correlations was observed between echocardiography indexes and TMA (total number = 11), butyric acid (total number = 10) and TMAO (total number = 9). Left atrium diameter index, which commonly increased in patients with atrial fibrillation, was correlated with TMA ($r=0.392$), TMAO ($r=0.333$), butyric acid ($r=-0.321$), isobutyric acid ($r=-0.359$) and middle chain fatty acids ($r=-0.362$), $P<0.05$.

Conclusion: Associations between gut microbiota metabolites and TTE AF characteristics in patients with CAD were based mostly on correlations between left atrium indexes and TMA, TMAO, butyric acid, isobutyric acid, and middle chain fatty acids.

Keywords: coronary artery disease, atrial fibrillation, echocardiography, gastrointestinal microbiome, fatty acids.

Corresponding author: *Iryna Melnychuk, Internal Medicine Department No. 4, Bogomolets National Medical University, Kyiv, Ukraine*
e-mail: ira.merkulova45@gmail.com

РЕЗЮМЕ

Ірина Мельничук
https://orcid.org/0000-0002-0659-1476
Кафедра внутрішньої медицини №4 НМУ імені О.О. Богомольця, м. Київ Україна

ЗВ'ЯЗКИ МЕТАБОЛІТІВ МІКРОБІОМУ КИШКІВНИКА З ЕХОКАРДІОГРАФІЧНИМИ ХАРАКТЕРИСТИКАМИ ФІБРИЛЯЦІЇ ПЕРЕДСЕРДЬ У ПАЦІЄНТІВ З ІШЕМІЧНОЮ ХВОРОБОЮ СЕРЦЯ

Мета: виявити зв'язок між метаболітами кишкової мікробіоти та ехокардіографічними (ЕхоКГ) характеристиками фібриляції передсердь (ФП) у пацієнтів з ішемічною хворобою серця (ІХС).

Матеріали і методи: 300 хворих було розподілено на 3 групи: перша (ІХС) – 149 пацієнтів з ІХС, але без аритмій, друга (ІХС+ФП) – 124 пацієнти з ІХС та пароксизмом ФП та контрольна група (КГ) – 27 пацієнтів без ІХС та аритмії. ЕхоКГ виконано ALOKA SSD-5000. Рівень триметиламіну (ТМА), триметиламін-N-оксиду (ТМАО) у плазмі та фекальних рівнях коротколанцюгових жирних кислот (КЖК) визначали за допомогою газової хроматографії з детекцією мас-електронів.

Результати: Ізокапронова та ізомасляна калові кислоти виявляються у II групі порівняно з КГ. У хворих II групи виявлено підвищення рівнів ТМА (16,13%), ТМАО (57,54%) та зниження їх співвідношення (26,16%) порівняно з I групою, P<0,05. У хворих II групи виявлено підвищення вмісту валеріанової (1128,43 %) та зниження масляної (78,75 %), ізовалеріанової (43,71 %), каприлової (99,21 %) кислот, жирних кислот середнього ланцюга (95,54 %) та загальної кількості КЖК калу (17,09 %) порівняно з I групою P<0,05. У хворих II групи відзначено збільшення діаметра лівого передсердя (10,03 %), об'єму лівого передсердя (15,40 %) та індексу об'єму лівого передсердя (11,48 %) порівняно з I групою, P<0,05. Найбільшу кількість кореляцій виявлено між ЕхоКГ показниками та ТМА (загальна кількість = 11), масляною кислотою (загальна кількість = 10) і ТМАО (загальна кількість = 9). Індекс діаметра лівого передсердя, який зазвичай підвищується у пацієнтів з фібриляцією передсердь, корелюється з ТМА (r=0,392), ТМАО (r=0,333), масляною кислотою (r=-0,321), ізомасляною кислотою (r=-0,359) та фекальними жирними кислотами середнього ланцюга (r=-0,362), P<0,05.

Висновок: зв'язок між метаболітами кишкової мікробіоти та характеристиками ЕхоКГ ФП у пацієнтів з ІХС базувався переважно на кореляції між індексами лівого передсердя та

ТМА, ТМАО, масляною кислотою, ізомасляною кислотою та жирними кислотами середнього ланцюга.

Ключові слова: ішемічна хвороба серця, фібриляція передсердь, ехокардіографія, шлунково-кишкова мікробіота, жирні кислоти.

Автор, відповідальний за листування *Iryna Melnychuk, кафедра внутрішньої медицини №4 НМУ імені О.О. Богомольця, м. Київ Україна*
e-mail: ira.merkulova45@gmail.com

ABBREVIATIONS

Trimethylamine (TMA); trimethylamine-N-oxide (TMAO); short-chain fatty acids (SCFA); lipopolysaccharide (LPS); cholesterol (TC); and low-density lipoproteins (LDL); atrial fibrillation (AF); coronary artery disease (CAD); transthoracic echocardiography (TTE); left atrium (LA); left ventricular (LV); ejection fraction (EF); control group (CG); myocardial infarction (MI); Glomerular Filtration Rate (GFR); body mass index (BMI); aorta diameter (AO); left ventricular outflow tract (LVOT); left atrium diameter (LAD); left atrium diameter index (LADI); left atrium volume (LAV); left atrium volume index (LAVI); right atrium diameter (RAD); right atrium diameter index (RADI); right atrium volume (RAV); right atrium volume index (RAVI); anterior-posterior size of the right ventricle (RV); interventricular septum (IVS); left ventricle posterior wall (LVPW); relative wall thickness (RWT); left ventricular mass (LVM); left ventricular mass index (LVMI); left ventricular stroke volume (LVSV); left ventricular stroke volume index (LVSVI); left ventricular end diastolic volume (LVEDV); left ventricular end diastolic volume index (LVEDVI); left ventricular end systolic volume (LVESV); left ventricular end systolic volume index (LVESVI); left ventricular end diastolic diameter (LVEDD); left ventricular end diastolic diameter index (LVEDDI); left ventricular end systolic diameter (LVESD); left ventricular end systolic diameter index (LVESDI); acetic acid (C2:0); propionic acid (C3:0); butyric acid (C4:0); isobutyric acid (C4:1); valeric acid (C5:0); isovaleric acid (C5:1); caproic acid (C6:0); isocaproic acid (C6:1); caprylic acid (C8:0); middle-chain fatty acids (MCFA); saturated fatty acids (SFA); unsaturated fatty acids (USFA); total amount of fecal SCFA (TA SCFA); confidence interval (CI).

INTRODUCTION / ВСТУП

Gut microbiota have been known to play crucial role in human health. Gut microbiota composition and function disturbances can disrupt gut barrier function and lead to metabolic endotoxemia. This low-grade chronic inflammation is an important basis in cardiometabolic events pathogenesis, such as dyslipidemia, obesity, atherosclerosis, thrombosis, insulin resistance, diabetes mellitus, aging, cancer, neurological and neurodegenerative disorders, etc. [1, 2]. Also, gut microbiota is involved in the synthesis of the majority of biologically active substances contributing to human health or disease development. The mean gut microbiota metabolites are trimethylamine (TMA) and trimethylamine-N-oxide (TMAO), short-chain fatty acids (SCFA), uremic toxins, bile acids, and lipopolysaccharide (LPS) [3].

Nowadays, TMAO is a known cardiometabolic risk factor. Higher TMAO concentrations increase the risk of cardiovascular events by 62%. TMAO is positively correlated with proinflammatory markers, oxidative stress molecules, cholesterol (TC), and low-density lipoproteins (LDL) [4]. Also, TMAO is associated with atrial fibrillation (AF) formation: activates atrial autonomic ganglion plexus and promotes arrhythmia, activates p65 nuclear factor-kB signaling, and increases expression of inflammatory cytokines. In animal studies rise of TMAO is associated with left ventricular hypertrophy and heart failure [1].

SCFA are a new potential contributor to AF pathogenesis. Low SCFA decreases gut integrity and leads to endotoxemia. SCFA decreases atrial remodeling, and have anti-inflammatory properties [1, 5]. According to some data, SCFA cardioprotective effect is based on regulatory T cell and T helper cell homeostasis, thereby reducing cardiac hypertrophy and fibrosis, atherosclerotic changes, that play an important role in cardiac arrhythmia pathogenesis. Moreover, SCFA inhibit NLRP3-inflammasome activity, which alters sarcoplasmic-reticulum Ca2+ release, exhibits ectopic activity, and shortened atrial effective refractory period, which leads to atrial hypertrophy [6].

AF and coronary artery disease (CAD) are widely spread cardiological conditions. They are the

independent risk factors of each other, which potentiate their development, worthier prognosis and complicate treatment. Furthermore, AF and CAD are associated with the same risk factors, such as dyslipidemia, obesity, inflammatory diseases, diabetes mellitus, arterial hypertension, aging, etc. [7, 8]. CAD and AF are characterized by special transthoracic echocardiography (TTE) peculiarities. AF is commonly associated with an increase in left atrium (LA) size and volume, LA dysfunction, left ventricular (LV) hypertrophy, LV systolic (decreased ejection fraction (EF)), and diastolic dysfunction [9, 10]. Besides, of importance and relevance of the investigation of associations between gut microbiota metabolites and TTE indexes in understanding the pathogenesis of AF paroxysm in CAD patients, this problem remains fully unexplored.

THE AIM: to reveal the connections between gut microbiota metabolites and echocardiography atrial fibrillation characteristics in patients with coronary artery disease.

MATERIALS AND METHODS

300 patients were enrolled in the study. They were divided into 3 groups: first – 149 patients with CAD but without arrhythmias, second – 124 patients with CAD and AF paroxysm, and the control group (CG) – 27 patients without CAD and arrhythmias. CAD and AF diagnoses were made according to the latest ESC guidelines [7, 8]. All patients were treated in the Kyiv City Clinical Hospital No. 12 in cardiological and therapeutic departments. AF paroxysm was checked by resting 12 leads electrocardiography. Diagnosis CAD was confirmed by a history of coronary arteries stenotic changes during invasive coronarography. All patients had heart failure stage B or C [11]. Exclusion criteria were: valvular AF, heart failure Class III to IV (by New York Heart Association), reported malignancies, thyroid pathology, chronic kidney disease (Glomerular Filtration Rate (GFR) < 60 mL/min), pregnancy, inflammatory bowel disease, irritable bowel syndrome, vegetarians and vegans, taking probiotics and antibiotics for a month before the study. A significant difference in risk factors at baseline was not seen between investigated groups. The study was conducted at the base and was approved by the ethical commission of the Kyiv City Clinical Hospital No. 12 (protocol # 8 from 22/08/2018). Informed consent was obtained from all subjects following the Declaration of Helsinki. Baseline characteristics of study patients include age, gender, history of myocardial infarction (MI), stroke, diabetes mellitus, obesity, body mass index (BMI), uric acid, total bilirubin, GFR, and TC levels. Uric acid, total bilirubin, creatinine, and TC were checked by the Kyiv City Clinical Hospital No. 12 laboratory (certificate # ПТ – 257/21). Advanced age, obesity, hypercholesterolemia, high stages of chronic kidney disease, gout, and hyperbilirubinemia are known risk factors of AF paroxysm development [7]. That's why these baseline characteristics were analyzed and compared because it can help us to exclude their influence on obtained results.

TTE was done by ALOKA SSD-5000. We analyzed such characteristics: aorta diameter (AO), left ventricular outflow tract (LVOT), left atrium diameter (LAD), left atrium diameter index (LADI), left atrium volume (LAV), left atrium volume index (LAVI), right atrium diameter (RAD), right atrium diameter index (RADI), right atrium volume (RAV), right atrium volume index (RAVI), anterior-posterior size of the right ventricle (RV), interventricular septum (IVS), left ventricle posterior wall (LVPW), relative wall thickness (RWT), left ventricular mass (LVM), left ventricular mass index (LVMI), EF, left ventricular stroke volume (LVSV), left ventricular stroke volume index (LVSVI), left ventricular end diastolic volume (LVEDV), left ventricular end diastolic volume index (LVEDVI), left ventricular end systolic volume (LVESV), left ventricular end systolic volume index (LVESVI), left ventricular end diastolic diameter (LVEDD), left ventricular end diastolic diameter index (LVEDDI), left ventricular end systolic diameter (LVESD), left ventricular end systolic diameter index (LVESDI) [9].

The level of TMAO, TMA plasma was determined by gas chromatography with mass electron detection. They were extracted from blood plasma into acid by adding internal standards [12]. The patient's blood sampling was performed on an empty stomach from the cubital vein on the day of hospitalization. Fecal SCFA was checked by gas chromatography with mass electron detection. We determined nine fatty acids in the collected samples – acetic acid (C2:0), propionic acid (C3:0), butyric acid (C4:0), isobutyric acid (C4:1), valeric acid (C5:0), isovaleric acid (C5:1), caproic acid (C6:0), isocaproic acid (C6:1) and caprylic acid (C8:0). These fatty acids include saturated (SFA) – acetic (C2:0), propionic (C3:0), butyric (C4:0), valeric (C5:0), caproic (C6:0), caprylic (C8:0) acids; and unsaturated (USFA) – isobutyric (C4:1), isovaleric (C5:1), isocaproic (C6:1) acids. Middle-chain fatty

acids (MCFA) include caproic acid (C6:0), isocaproic acid (C6:1), and caprylic acid (C8:0). Also, the total amount of fecal SCFA (TA SCFA) was determined [13].

Results were presented as mean ± standard error or [95% confidence interval (CI)] for continuous variables or as a number for categorical variables. Variables distribution for normality were checked by the Pearson criterion. Data were compared using the Wilcoxon signed-rank test or Student t-test with two critical regions by the type of distribution; Spearman's rank correlation coefficient [14]. All calculations were done in MATLAB R2014a (License number 271828).

RESULTS

Firstly, baseline characteristics of investigated groups were analyzed. Significant difference in age, gender, BMI, total bilirubin, and smoking history was not found in investigated groups. In groups I and II uric acid (by 22.66% and 30.53% respectively) and TC (by 32.64% and 43.06% respectively) levels were significantly higher and GFR (by 26.16% and 19.38% respectively) were lower than in CG ($p<0.05$). Also, in groups I and II there were patients with obesity, diabetes mellitus, stroke, or MI history, such cases were absent in CG. Data are shown in Table 1.

Table 1 – Baseline characteristics of study groups, mean ± standard error

Characteristic /group	I group	II group	CG	P1-2	P2-CG	P1-CG
Age (years)	67.71±3.90	67.96±0.94	56.25±2.18	P>0.05	P>0.05	P>0.05
Men (%)	48.99	47.97	48.15	P>0.05	P>0.05	P>0.05
Smoking (%)	51.01	41.46	40.74	P>0.05	P>0.05	P>0.05
History of myocardial infarction (%)	30.87	26.02	0	P>0.05	P<0.05	P<0.05
History of stroke (%)	8.72	8.13	0	P>0.05	P<0.05	P<0.05
Diabetes mellitus (%)	18.12	14.63	0	P>0.05	P<0.05	P<0.05
Obesity (%)	8.84	12.0	0	P>0.05	P<0.05	P<0.05
BMI (kg/m^2)	27.02±0.33	26.93±0.43	27.12±2.10	P>0.05	P>0.05	P>0.05
Total bilirubin (mmol/l)	11.3±0.09	12.4±0.08	11.7±0.11	P>0.05	P>0.05	P>0.05
Uric acid (mmol/l)	380.5±28.16	404.9±36.11	310.2±29.12	P>0.05	P<0.05	P<0.05
GFR (ml/min)	62.03±2.31	67.73±1.98	84.01±5.48	P>0.05	P<0.05	P<0.05
TC (mmol/l)	5.73±0.37	6.18±0.31	4.32±0.21	P>0.05	P<0.05	P<0.05

Secondary, TMA, TMAO, and their ratio levels were investigated. In groups II and I, we found a significant rise of TMA (22.50% and 42.25% respectively), TMAO (50.00% and 136.31% respectively) levels and decrease TMA/TMAO ratio (18.59% and 39.89% respectively) in comparison with CG, $P<0.05$. Also, in group II in comparison with group I significant rise of TMA (16.13%), and TMAO (57.54%) levels, and a decrease in TMA/TMAO ratio (26.16%) was revealed, $P<0.05$. Results are shown in Figure 1.

Also, we investigated fecal SCFA composition. Due to the obtained data in group I was a significant rise of isovaleric acid (165.58%), USFA (485.44%) and decrease of butyric (63.36%), valeric (97.75%), caproic (93.39%) acids, TA SCFA (29.52%%), MCFA (66.04%) in comparison with CG, $P<0.05$. In group II was found significant rise of acetic (62.35%) and a decrease of butyric (92.21%), valeric (72.36%), caprylic (99.84%) acids, TA SCFA (41.57%) in comparison with CG, $P<0.05$. Besides, in group II was a significant rise of valeric (1128.43%) and depletion of butyric (78.75%), isovaleric (43.71%), caprylic (99.21%) acids, MCFA (95.54%), and TA SCFA (17.09%) in comparison with group I, $P<0.05$. Isocaproic and isobutyric fecal acids were not found in the CG samples, but they appeared in group I and II patients' tests. Results are shown in Figures 2 and 3.

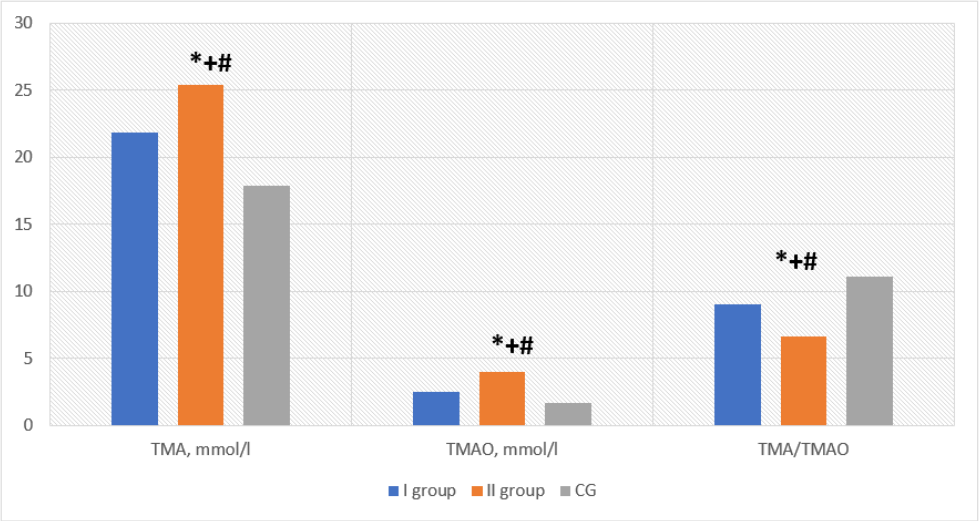

Figure 1 – TMA, TMAO, and their ratio in investigated groups, mmol/l
*Notes: * – P<0.05 I-II groups; + – P<0.05 I group – CG; # – P<0.05 II group – CG*

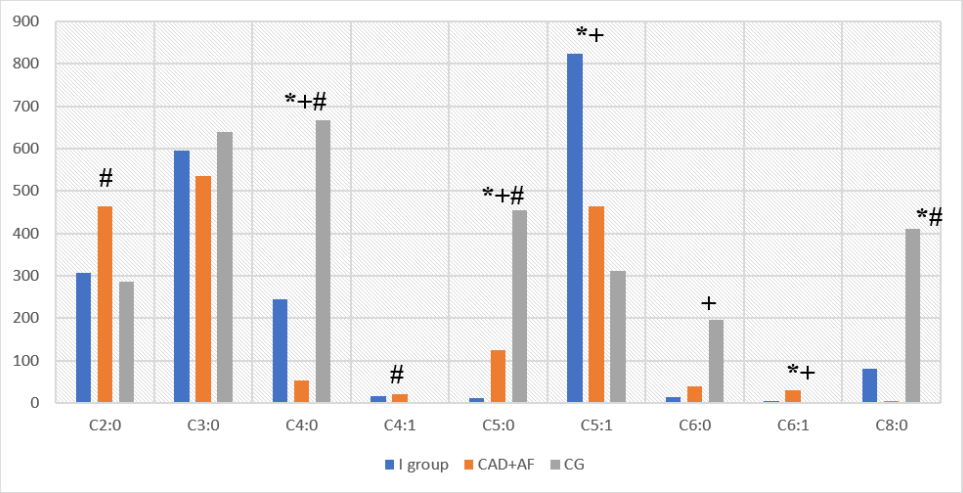

Figure 2 – Fecal short-chain fatty acids in investigated groups, mg/g
*Notes: * – P<0.05 I-II groups; + – P<0.05 I group – CG; # – P<0.05 II group – CG*

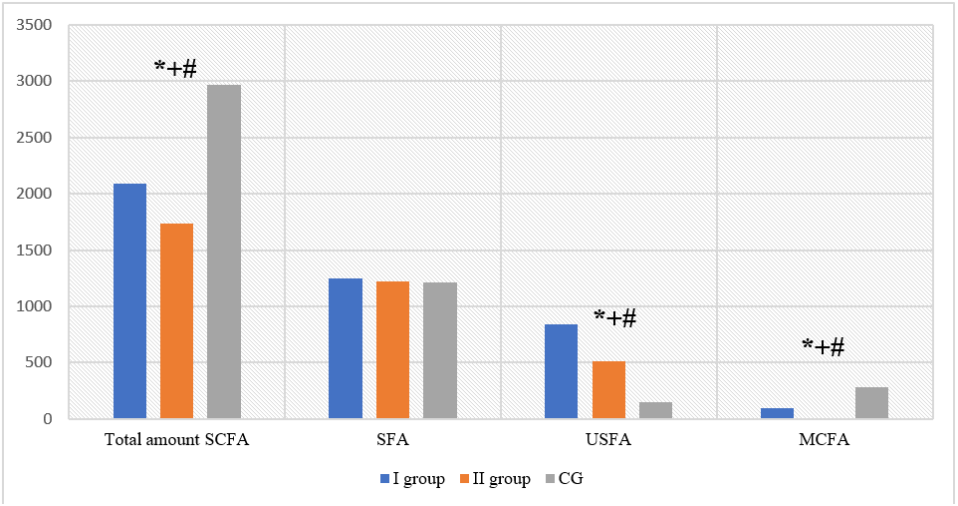

Figure 3 – Fecal short-chain fatty acids in investigated groups, mg/g
*Notes: * – P<0.05 I-II groups; + – P<0.05 I group – CG; # – P<0.05 II group – CG*

TTE indexes were explored in all investigated groups. In group I were significant increases in IVS (15.15%), LVPW (17.71%), RWT (12.35%), LVM (39.78%), LVMI (35.87%), LVEDV (21.42%) in comparison with CG. In group II were significant rise of LVOT (11.47%), LAD (10.03%), LAV (15.40%), LAVI (11.48%), RV (11.50%), IVS (18.18%), LVPW (20.83%), LVM (47.93%), LVMI (46.02%), LVEDV (27.28%), LVEDVI (25.88%) in comparison with CG. Also, in group II significant increases in LAD (10.03%), LAV (15.40%), and LAVI (11.48%) were found in comparison with group I ($p < 0.05$). Data are presented in Table 2.

Table 2 – TTE of investigated groups, mean ± standard error

Characteristic /group	I group	II group	CG	P1-2	P2-CG	P1-CG
AO, cm	3.19±0.04	3.27±0.04	3.06±0.06	P>0.05	P>0.05	P>0.05
LVOT, cm	3.05±0.03	3.11±0.03	2.79±0.09	P>0.05	P<0.05	P>0.05
LAD, cm	4.08±0.04	4.28±0.05	3.89±0.06	P<0.05	P<0.05	P>0.05
LADI, cm/m^2	2.12±0.04	2.26±0.04	2.08±0.05	P>0.05	P>0.05	P>0.05
LAV, ml	3.95±0.03	4.42±0.05	3.83±0.04	P<0.05	P<0.05	P>0.05
LAVI, ml/m^2	2.05±0.03	2.33±0.04	2.09±0.03	P<0.05	P<0.05	P>0.05
RAD, cm	3.52±0.04	3.65±0.04	3.48±0.05	P>0.05	P>0.05	P>0.05
RADI, cm/m^2	1.83±0,03	1,92±0,03	1,87±0,04	P>0.05	P>0.05	P>0.05
RAV, ml	3.85±0.03	3.94±0.03	3.94±0.03	P>0.05	P>0.05	P>0.05
RAVI, ml/m^2	2.00±0.02	2.11±0.03	2.11±0.03	P>0.05	P>0.05	P>0.05
RV, cm	2.48±0.03	2.52±0.03	2.26±0.02	P>0.05	P<0.05	P>0.05
IVS, cm	1.14±0.01	1.17±0.02	0.99±0.01	P>0.05	P<0.05	P<0.05
LVPW, cm	1.13±0.01	1.16±0.01	0.96±0.02	P>0.05	P<0.05	P<0.05
RWT	0.91±0.02	0.96±0.02	0.81±0.04	P>0.05	P>0.05	P<0.05
LVM, g	188.70±5.60	199.70±7.26	135.00±6.01	P>0.05	P<0.05	P<0.05
LVMI, g/m^2	98.07±3.26	105.40±3.97	72.18±3.39	P>0.05	P<0.05	P<0.05
EF	0.59±0.01	0.59±0.01	0.59±0.01	P>0.05	P>0.05	P>0.05
LVSV, ml	57.67±1.59	59.93±1.90	48.24±3.61	P>0.05	P>0.05	P>0.05
LVSVI, ml/m^2	29.83±1.04	31.64±1.24	25.88±2.01	P>0.05	P>0.05	P>0.05
LVEDV, ml	98.64±2.77	103.40±3.73	81.24±4.62	P>0.05	P<0.05	P<0.05
LVEDVI, ml/m^2	51.37±1.85	54.87±2.34	43.59±2.62	P>0.05	P<0.05	P>0.05
LVESV, ml	40.97±1.60	43.46±2.47	33.00±1.59	P>0.05	P>0.05	P>0.05
LVESVI, ml/m^2	21.54±1.04	23.24±1.53	17.71±0.92	P>0.05	P>0.05	P>0.05
LVEDD, cm	4.50±0.06	4.61±0.06	4.23±0.09	P>0.05	P>0.05	P>0.05
LVEDDI, cm/m^2	2.33±0.04	2.40±0.06	2.26±0.06	P>0.05	P>0.05	P>0.05
LVESD, cm	3.05±0.05	3.15±0.06	2.99±0.10	P>0.05	P>0.05	P>0.05
LVESDI, cm/m^2	1.59±0.03	1.64±0.05	1.60±0.06	P>0.05	P>0.05	P>0.05

The correlation analysis between gut microbiota metabolites and TTE indexes was done in the investigated groups. Spearman's correlation analysis was used to explore their correlations with species abundance. The largest amount of correlations was checked between echocardiography indexes and TMA (total number = 11), butyric acid (total number = 10) and TMAO (total number = 9). Also, the

highest amount of correlations were found between LADI (total number = 5), EF (total number = 5), RWT (total number = 6), and gut microbiota metabolites. TMA was correlated with LAD (r=0.326), LADI (r=0.392), LAV (r=0.395), LAVI (r=0.454), RADI (r=0.393), RAVI (r=0.423), LVM (r=0.362), LVMI (r=0.323), LVEDV (r=0.347), LVEDVI (r=0.447) and LVEDD (r=0.314), P<0.05. TMAO was corelated with LADI (r=0.333), LAV (r=0.341), LAVI (r=0.361), RADI (r=0.366), RAVI (r=0.435), LVMI (r=0.364), EF (r=-0.382), LVEDVI (r=0.348) and LVESVI (r=0.414), P<0.05. Butyric acid was correlated with LADI (r=-0.321), LAV (r=-0.355), LAVI (r=-0.476), RADI (r=-0.310), RAVI (r=-0.314), LVMI (r=-0.303), LVSV (r=-0.335), LVSVI (r=-0.446) and LVEDVI (r=-0349), P<0.05. LADI was correlated with TMA (r=0.392), TMAO (r=0.333), butyric acid (r=-0.321), isobutyric acid (r=-0.359), and MCFA (r=-0.362), P<0.05. All correlations are shown in the Figure 4.

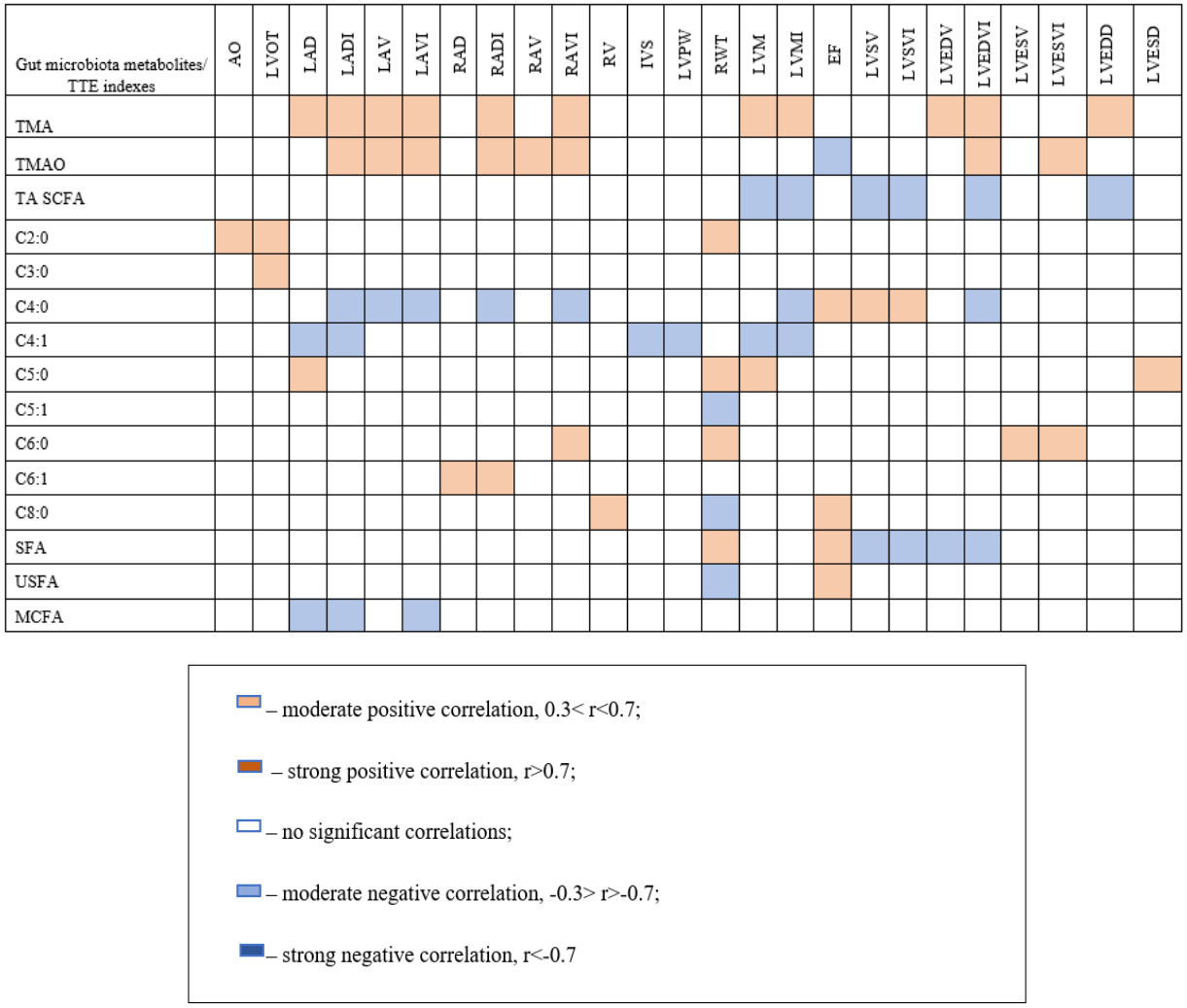

Figure 3 – Heatmap correlation matrices between gut microbiota metabolites and echocardiography indexes, P<0.05

DISCUSSION

Due to our results associations between gut microbiota metabolites and TTE AF characteristics in patients with CAD was based mostly on correlations between left atrium indexes and plasma TMA, TMAO, butyric acid, isobutyric acid and middle chain fatty acids. It is widely known that patients with AF are characterized by an increase in left atrium indexes: LAD, LADI, LAV, and LAVI. It is the reliable marker of LA structural remodeling, which is associated with the development of AF and its complications [9, 10]. That LA structural remodeling (laid down) is based on processes of cardiac fibrosis, cell apoptosis and connection lateralization [1].

Nowadays TMA and TMAO are the most commonly investigated gut microbiota metabolites. It is closely linked with atherosclerosis, coronary

artery disease, arterial hypertension, diabetes mellitus, obesity, and another metabolic dysfunction pathogenesis. Moreover, several studies have suggested that TMAO is inextricably linked with the occurrence, progression, and recurrence of AF after ablation [1]. Although there are the majority of relevant studies, specific mechanisms of AF induction by TMAO are still under discussion. It increased neural activity and the susceptibility of atrial to AF through the p65 NF-κB signaling pathway. Also, TMAO induces cardiac hypertrophy and fibrosis via the Smad3 signaling pathway. Besides, TMAO potentiates cardiac fibrosis via activation of the NLRP3 inflammasome and activates oxidative stress, by the activation of the p53/p21/Rb pathway. Undoubtedly, all this is connected with LA structural, autonomic, and electrical remodeling [1, 15].

SCFA are produced by gut microbiota during glucose and dietary fiber fermentation. SCFA improves intestinal barrier functions by promoting mucous production, affects immune regulation by inhibiting histone deacetylases, and alleviates NLRP3 signaling-mediated atrial remodeling. So, lack of fecal SCFA is associated with Ca2+ handling and structural LA remodeling [1, 16]. Moreover, SCFA differ in their properties. Butyrate has a strong anti-inflammatory effect, increases insulin sensitivity, and normalizes lipid exchange. In the animal studies, butyrate showed its protection effects for LA electrical remodeling (action potential duration shortening, L-type Ca^{2+}-current reduction), cellular Ca^{2+}-handling/contractile dysfunction, and endoplasmic reticulum stress and autophagy [1, 17]. Vice versa, acetate promotes parasympathetic nervous system activation by the brain and increases insulin and ghrelin secretion leading to obesity and dyslipidemia [1]. Today there is a lack of data about the role of MCFA in human health. Due to in vitro studies, caproic acid has postbiotic properties, it inhibits pathogenic flora (for example, *E. coli*), so has strong antimicrobial properties and antioxidant activity [18]. Caprylic acid reduces inflammation (decreases interleukine-1β, interleukine-6, tumor necrotic factor-α, and monocyte chemoattractant protein-1 levels) and normalizes lipids exchange (decreases triglycerides and increases high-density lipoprotein levels) in animal studies [19]. So, SCFA metabolism is also closely connected with AF risk factors, thereby LA structural remodeling.

Nevertheless, the presence of active scientific interest in the role of gut microbiota metabolites in hemodynamic changes in CAD and AF patients, this problem still has not been resolved yet.

CONCLUSIONS / ВИСНОВКИ

During our study peculiarities of gut microbiota metabolites profile and echocardiography indexes and their associations in patients with coronary artery disease and atrial fibrillation were revealed:

1. Isocaproic and isobutyric fecal acids appeared in coronary artery disease and atrial fibrillation patients' samples in comparison with the control group;
2. In the patients with atrial fibrillation and coronary artery disease, a rise of trimethylamine (16.13%), and trimethylamine-N-oxide (57.54%) levels and decreased their ratio (26.16%) in comparison with coronary artery disease patients without arrhythmia was revealed, $P<0.05$;
3. In the patients with atrial fibrillation and coronary artery disease rise of valeric (1128.43%) and depletion of butyric (78.75%), isovaleric (43.71%), caprylic (99.21%) acids, middle chain fatty acids (95.54%) and total amount fecal short chain fatty acids (17.09%) in comparison with coronary artery disease patients without arrhythmia was found, $P<0.05$;
4. In the patients with atrial fibrillation and coronary artery disease increase of left atrium diameter (10.03%), left atrium volume (15.40%), and left atrium volume index (11.48%) in comparison with coronary artery disease patients without arrhythmia was established, $P<0.05$;
5. The largest amount of positive and negative correlations were found between echocardiography indexes and TMA (total number = 11), butyric acid (total number = 10), and TMAO (total number = 9);
6. Left atrium diameter index, which commonly increased in patients with atrial fibrillation, was strongly correlated with TMA ($r=0.392$), TMAO ($r=0.333$), butyric acid ($r=-0.321$), isobutyric acid ($r=-0.359$) and MCFA ($r=-0.362$), $P<0.05$.

PROSPECTS FOR FUTURE RESEARCH / ПЕРСПЕКТИВИ ПОДАЛЬШИХ ДОСЛІДЖЕНЬ

Other gut microbiota metabolites and TTE indexes connections investigations will be interesting for further scientific studies.

CONFLICT OF INTEREST / КОНФЛІКТ ІНТЕРЕСІВ

The author declares that there is no conflict of interest regarding this research, including financial, personal, authorship, or other nature, which could affect the research and its results presented in this article.

FUNDING / ДЖЕРЕЛА ФІНАНСУВАННЯ

This study did not receive external funding. The study was done according to the department scientific study work "Changes in protein, carbohydrate and lipid metabolism in patients with coronary heart disease and arterial hypertension with heart rhythm disorders, possibilities of drug correction" 2021-2023 (state registration number 0121U108875).

AUTHOR CONTRIBUTIONS / ВКЛАД АВТОРІВ

Conception, methodology, data collection, analysis, review, writing, and administration – Melnychuk I. O.

REFERENCES/СПИСОК ЛІТЕРАТУРИ

1. Gawałko M, Agbaedeng TA, Saljic A, Müller DN, Wilck N, Schnabel R, Penders J, Rienstra M, van Gelder I, Jespersen T, Schotten U, Crijns HJGM, Kalman JM, Sanders P, Nattel S, Dobrev D, Linz D. Gut microbiota, dysbiosis and atrial fibrillation. Arrhythmogenic mechanisms and potential clinical implications. *Cardiovasc Res.* 2022 Aug 24;118(11):2415-2427. https://doi.org/10.1093/cvr/cvab292. PMID: 34550344; PMCID: PMC9400433.
2. Patterson E, Ryan PM, Cryan JF, Dinan TG, Ross RP, Fitzgerald GF, Stanton C. Gut microbiota, obesity and diabetes. *Postgrad Med J.* 2016 May;92(1087):286-300. https://doi.org/10.1136/postgradmedj-2015-133285. Epub 2016 Feb 24. PMID: 26912499.
3. Wang Z, Zhao Y. Gut microbiota derived metabolites in cardiovascular health and disease. *Protein Cell.* 2018 May;9(5):416-431. https://doi.org/10.1007/s13238-018-0549-0. Epub 2018 May 3. PMID: 29725935; PMCID: PMC5960473.
4. Swanepoel I, Roberts A, Brauns C, Chaliha DR, Papa V, Palmer RD, Vaccarezza M. Trimethylamine N-oxide (TMAO): a new attractive target to decrease cardiovascular risk. *Postgrad Med J.* 2022 Sep;98(1163):723-727. https://doi.org/10.1136/postgradmedj-2021-139839. Epub 2021 Mar 31. PMID: 33790031.
5. Chen L, Chen J, Huang Y, Wu Y, Li J, Ni W, Lu Y, Li Z, Zhao C, Kong S, Zhou H, Qu X. Changes of the gut microbiota composition and short chain fatty acid in patients with atrial fibrillation. *PeerJ.* 2023 Dec 7;11:e16228. https://doi.org/10.7717/peerj.16228. PMID: 38084144; PMCID: PMC10710774.
6. Zhang J, Zuo K, Fang C, Yin X, Liu X, Zhong J, Li K, Li J, Xu L, Yang X. Altered synthesis of genes associated with short-chain fatty acids in the gut of patients with atrial fibrillation. *BMC Genomics.* 2021 Aug 31;22(1):634. https://doi.org/10.1186/s12864-021-07944-0. PMID: 34465304; PMCID: PMC8406843.
7. Hindricks G, Potpara T, Dagres N, Arbelo E, Bax JJ, Blomström-Lundqvist C, Boriani G, Castella M, Dan GA, Dilaveris PE, Fauchier L, Filippatos G, Kalman JM, La Meir M, Lane DA, Lebeau JP, Lettino M, Lip GYH, Pinto FJ, Thomas GN, Valgimigli M, Van Gelder IC, Van Putte BP, Watkins CL. Corrigendum to: 2020 ESC Guidelines for the diagnosis and management of atrial fibrillation developed in collaboration with the European Association for Cardio-Thoracic Surgery (EACTS): The Task Force for the diagnosis and management of atrial fibrillation of the European Society of Cardiology (ESC) Developed with the special contribution of the European Heart Rhythm Association (EHRA) of the ESC. *Eur Heart J.* 2021 Oct 21;42(40):4194. https://doi.org/10.1093/eurheartj/ehab648. Erratum for: Eur Heart J. 2021 Feb 1;42(5):373-498. PMID: 34520521.
8. Knuuti J, Wijns W, Saraste A, Capodanno D, Barbato E, Funck-Brentano C, Prescott E, Storey RF, Deaton C, Cuisset T, Agewall S, Dickstein K, Edvardsen T, Escaned J, Gersh BJ, Svitil P, Gilard M, Hasdai D, Hatala R, Mahfoud F, Masip J, Muneretto C, Valgimigli M, Achenbach S, Bax JJ; ESC Scientific Document Group. 2019 ESC Guidelines for the diagnosis and management of chronic coronary syndromes. *Eur Heart J.* 2020 Jan 14;41(3):407-477. https://doi.org/10.1093/eurheartj/ehz425. Erratum in: Eur Heart J. 2020 Nov 21;41(44):4242. PMID: 31504439.
9. Mitchell C, Rahko PS, Blauwet LA, Canaday B, Finstuen JA, Foster MC, Horton K, Ogunyankin KO, Palma RA, Velazquez EJ. Guidelines for Performing a Comprehensive Transthoracic Echocardiographic Examination in Adults: Recommendations from the American Society of Echocardiography. *J Am Soc Echocardiogr.* 2019 Jan;32(1):1-64.

https://doi.org/10.1016/j.echo.2018.06.004. Epub 2018 Oct 1. PMID: 30282592.

10. Tufano A, Galderisi M. Can echocardiography improve the prediction of thromboembolic risk in atrial fibrillation? Evidences and perspectives. *Intern Emerg Med.* 2020 Sep;15(6):935-943. https://doi.org/10.1007/s11739-020-02303-5. Epub 2020 Mar 2. PMID: 32124208.
11. Authors/Task Force Members:; McDonagh TA, Metra M, Adamo M, Gardner RS, Baumbach A, Böhm M, Burri H, Butler J, Čelutkienė J, Chioncel O, Cleland JGF, Crespo-Leiro MG, Farmakis D, Gilard M, Heymans S, Hoes AW, Jaarsma T, Jankowska EA, Lainscak M, Lam CSP, Lyon AR, McMurray JJV, Mebazaa A, Mindham R, Muneretto C, Francesco Piepoli M, Price S, Rosano GMC, Ruschitzka F, Skibelund AK; ESC Scientific Document Group. 2023 Focused Update of the 2021 ESC Guidelines for the diagnosis and treatment of acute and chronic heart failure: Developed by the task force for the diagnosis and treatment of acute and chronic heart failure of the European Society of Cardiology (ESC) With the special contribution of the Heart Failure Association (HFA) of the ESC. *Eur J Heart Fail.* 2024 Jan 3. https://doi.org/10.1002/ejhf.3024. Epub ahead of print. PMID: 38169072.
12. Bain MA, Faull R, Fornasini G, Milne RW, Schumann R, Evans AM. Quantifying trimethylamine and trimethylamine-N-oxide in human plasma: interference from endogenous quaternary ammonium compounds. *Anal Biochem.* 2004 Nov 15;334(2):403-5. https://doi.org/10.1016/j.ab.2004.07.004. PMID: 15494149.
13. Michelle SW. Xiang, Jian K. Tan, Laurence Macia Fatty Acids, Gut Bacteria, and Immune Cell Function. *The Molecular Nutrition of Fats*, 2019; 11:151-164. https://doi.org/10.1016/B978-0-12-811297-7.00011-1.
14. Faizi N, Alvi Y. *Biostatistics Manual for Health Research.* Elsevier; 2023. 290 p.
15. Huang R, Yan L, Lei Y. The Gut Microbial-Derived Metabolite Trimethylamine N-Oxide and Atrial Fibrillation: Relationships, Mechanisms, and Therapeutic Strategies. *Clin Interv Aging*. 2021 Nov 30;16:1975-1986. https://doi.org/10.2147/CIA.S339590. PMID: 34876810; PMCID: PMC8643130.
16. Zuo K, Fang C, Liu Z, Fu Y, Liu Y, Liu L, Wang Y, Yin X, Liu X, Li J, Zhong J, Chen M, Xu L, Yang X. Commensal microbe-derived SCFA alleviates atrial fibrillation via GPR43/NLRP3 signaling. *Int J Biol Sci.* 2022 Jun 27;18(10):4219-4232. https://doi.org/10.7150/ijbs.70644. PMID: 35844801; PMCID: PMC9274492.
17. Wiersma M, Meijering RAM, Qi XY, Zhang D, Liu T, Hoogstra-Berends F, Sibon OCM, Henning RH, Nattel S, Brundel BJJM. Endoplasmic Reticulum Stress Is Associated With Autophagy and Cardiomyocyte Remodeling in Experimental and Human Atrial Fibrillation. *J Am Heart Assoc.* 2017 Oct 24;6(10):e006458. https://doi.org/10.1161/JAHA.117.006458. PMID: 29066441; PMCID: PMC5721854.
18. Chang HM, Foo HL, Loh TC, Lim ETC, Abdul Mutalib NE. Comparative Studies of Inhibitory and Antioxidant Activities, and Organic Acids Compositions of Postbiotics Produced by Probiotic *Lactiplantibacillus plantarum* Strains Isolated From Malaysian Foods. *Front Vet Sci.* 2021 Jan 26;7:602280. https://doi.org/10.3389/fvets.2020.602280. PMID: 33575277; PMCID: PMC7870707.

Received 09.01.2024
Accepted 30.01.2024

Одержано 09.01.2024
Затверджено до друку 30.01.2024

INFORMATION ABOUT THE AUTHORS / ВІДОМОСТІ ПРО АВТОРІВ

Melnychuk Iryna, PhD, Associate Professor of Internal Medicine Department No. 4, Bogomolets National Medical University, bulv. Shevchenko 13, Kyiv, Ukraine, 01030, tel. +380502893355, email: ira.merkulova45@gmail.com, ORCID ID: https://orcid.org/0000-0002-0659-1476