



*colloquium-journal*

**ISSN 2520-6990**

**Międzynarodowe czasopismo naukowe**

**Earth sciences  
Medical sciences  
Technical science  
Biological sciences  
Agricultural sciences  
Physics and mathematics**

**№32(119) 2021**

**Część 1**



*colloquium-journal*

ISSN 2520-6990

ISSN 2520-2480

Colloquium-journal №32 (119), 2021

Część 1

(Warszawa, Polska)

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### МЕХАНІЗМИ ВЗАЄМОБТЯЖЕННЯ НЕАЛКОГОЛЬНОЇ ЖИРОВОЇ ХВОРОБИ ПЕЧІНКИ ТА ХРОНІЧНОЇ ХВОРОБИ НИРОК

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### THE MECHANISMS OF INTERACTION OF NON-ALCOHOLIC FATTY LIVER DISEASE AND CHRONIC KIDNEY DISEASE

#### Анотація.

За результатами нашого дослідження було встановлено, що найбільш суттєвими метаболічними передумовами розвитку неалкогольної жирової хвороби печінки на тлі ожиріння та хронічної хвороби нирок є вірогідна постпрандіальна гіперглікемія, гіперінсулінемія та зростання ступеня глікозилювання гемоглобіну. Причиною прогресування метаболічного синдрому на тлі неалкогольної жирової хвороби печінки та хронічної хвороби нирок є ліпідний дистрес-синдром із зростанням у крові проатерогенних ліпопротеїнів низької щільності, дефіцитом антиатерогенних ліпопротеїнів високої щільності. Провідну роль у розвитку та прогресуванні неалкогольного стеатогепатиту та розладів печінкового кровообігу справляє зростання в крові тригліцеридів. Таким чином, розвиток неалкогольної жирової хвороби печінки у пацієнтів з хронічною хворобою нирок та ожирінням супроводжується суттєвою дис- та гіперліпідемією із максимальним серед груп порівняння зростанням вмісту в крові холестеролу та проатерогенних ліпопротеїнів низької щільності, вірогідним зниженням протиатерогенних ліпопротеїнів високої щільності та зростанням індексу атерогенності.

#### Summary.

The study showed that the most significant metabolic prerequisites for the development of nonalcoholic fatty liver disease on the background of obesity and chronic kidney disease are probable postprandial hyperglycemia, hyperinsulinemia and increased glycosylation of hemoglobin. The reason for the progression of the metabolic syndrome on the background of non-alcoholic fatty liver disease and chronic kidney disease is lipid distress syndrome with an increase in blood low-density proatherogenic lipoproteins, deficiency of high-density antiatherogenic lipoproteins. The leading role in the development and progression of non-alcoholic steatohepatitis and hepatic circulatory disorders is played by the growth of triglycerides in the blood. Thus, the development of non-alcoholic fatty liver disease in patients with chronic kidney disease and obesity is accompanied by significant dys- and hyperlipidemia with the maximum among the groups of comparison increase in blood cholesterol and low-density proatherogenic lipoproteins, probable decrease in antiatherogenesis and atherogenesis.

**Ключові слова:** неалкогольна жирова хвороба печінки, хронічна хвороба нирок, ожиріння.

**Keywords:** non-alcoholic fatty liver disease, chronic kidney disease, obesity.

**Introduction.** An important problem of internal medicine is the problem of comorbidity of non-alcoholic fatty liver disease (NAFLD) with obesity, chronic kidney disease (CKD), which has significant general medical and social significance [1, 2]. The range of diseases included in the concept of NAFLD includes non-alcoholic hepatic steatosis (NASP), steatohepatitis (NASH), which may be accompanied by liver fibrosis (AF) and transform into liver cirrhosis (CP). The prevalence of NAFLD in the population is 10-33%. The prevalence of NASH in the world is 10% (600 million people) [4, 5, 8]. Over the last 5 years, the incidence of steatohepatitis in Ukraine has increased by 76.6%. 12-40% of patients with hepatic steatosis develop NASH with early AF within 8-13 years. 25% of them develop

CP, hepatocellular insufficiency (PKN) (15%) or precirrotic changes (10%). 7% of patients with compensated CP develop hepatocellular carcinoma within 10 years, and 50% of them require liver transplantation or die from PKN. kidney disease, obesity.

**The purpose of the study:** to determine the intensity of the mechanisms of mutual burdening of non-alcoholic fatty liver disease on the background of obesity, depending on its form in the presence of comorbid chronic kidney disease and its stage.

**Material and methods of research.** 135 patients with non-alcoholic steatohepatitis (NASH) with comorbid obesity I degree and chronic kidney disease (CKD) of the 1st and 2nd stage, were examined. Patients were divided into 2 groups: of which 52 patients

with non-alcoholic steatohepatitis with obesity I degree (group 1), 53 patients with non-alcoholic steatohepatitis with comorbid obesity of the 1st degree and chronic kidney disease of the I-II stage (chronic uncomplicated pyelonephritis with latent phase in subsiding exacerbation phase) (group 2). The control group consisted of 30 practically healthy persons of the corresponding age and sex. The average age of patients was  $(45.8 \pm 3.81)$  years, men were 48, and women 57 persons. The functional state of the endothelium was studied by the content of stable metabolites of nitrogen monoxide (NO) (nitrites, nitrates) in the blood by L.C.Green et al. The number of desquamated endothelial cells in the blood was determined by the method of J.Hladovec in the modification of N.N. Petrishev et al. The lipid blood spectrum was studied based on the content of common lipids (TL), total cholesterol, triacylglycerols (TG), low density lipoprotein (LDL) and high-density lipoprotein (HDL) (Danish Ltd, Lviv), and also calculated the index of atherogenicity (IA) by the formula:  $IA = \text{total cholesterol} / \text{HDL}$ . The degree of carbohydrate compensation was determined by the level of glycemia in the onset and 2 hours after glucose loading (glucose tolerance test) by the glucose oxidase method, the content of insulin in the blood (DRG System) - by the immunoassay (ELISA) method, the content of glycosylated hemoglobin (HbA1c) using standard sets of reagents (DanishLtd, Lviv) by the method of V.A. Koroleva.

The diagnosis of NASH was established in accordance with the unified clinical protocol, approved by the order of the Ministry of Health of Ukraine No. 826 from 06.11.2014, in the presence of criteria for the exclusion of chronic diffuse liver disease of the viral, hereditary,

autoimmune or medicinal genesis as causes of cholestatic or cytolytic syndromes, as well as the results of the USG survey. Diagnosis and treatment of CKD were performed according to the recommendations of the clinical guidelines of the State Institute "Institute of Nephrology, NAMS of Ukraine" (2012).

The statistical analysis of the results was carried out in accordance with the type of research carried out and the types of numerical data that were obtained. Distribution normality was verified using Liliefors, Shapiro-Uilka tests and the direct visual evaluation of eigenvalues distribution histograms. Quantitative indices having a normal distribution are represented as mean (M)  $\pm$  standard deviation (S). Discrete values are presented in the form of absolute and relative frequencies (percentage of observations to the total number of surveyed). For comparisons of data that had a normal distribution pattern, parametric tests were used to estimate the Student's t-criterion, Fisher's F-criterion. In the case of abnormal distribution, the median test, Mann-Whitney Rank U-Score, and Wilcoxon's T-criterion (in the case of dependent groups) were used for multiple comparison. Statistica for Windows version 8.0 (Stat Soft inc., USA), Microsoft Excel 2007 (Microsoft, USA) software packages were used for statistical and graphical analysis of the obtained results.

#### Results of the research and their discussion.

Analysis of the lipid profile of the blood in patients with NASH and obesity showed a number of changes that differed depending on the presence of CKD (**table**). Indicators of concentration in blood of total lipids in patients of 1st and 2nd groups exceeded the norm by 26.4% and 34.2%, respectively, with a statistically significant difference between the groups ( $p < 0.05$ ).

Table

#### Indicators of lipid spectrum of blood and endothelial dysfunction in patients with non-alcoholic stethohepatitis, obesity of the I-II degree and with comorbidity with chronic kidney disease of the I-II stage (M $\pm$ m)

Indicators, units measurement	Groups of patients surveyed		
	PHP	Group 1 NASH+Obesity	Group 2 NASH with CKD + Obesity
Total Cholesterol, mmol / l	4,72 $\pm$ 0,11	6,89 $\pm$ 0,38*	6,93 $\pm$ 0,39*/**
LDL, mmol / l	2,54 $\pm$ 0,02	4,05 $\pm$ 0,022 *	4,58 $\pm$ 0,04*/**
HDL, mmol / l	1,28 $\pm$ 0,05	0,72 $\pm$ 0,02 *	0,76 $\pm$ 0,04 */**
TG, mmol / l	1,47 $\pm$ 0,03	2,42 $\pm$ 0,03*	3,19 $\pm$ 0,07 */**
NO IN BLOOD, mmol / l	15,32 $\pm$ 1,225	30,49 $\pm$ 1,318 *	40,51 $\pm$ 1,173 */**
ET-1, pmol / l	6,17 $\pm$ 0,854	11,25 $\pm$ 0,457 *	18,83 $\pm$ 0,559 */**
DEC x104/L	3,03 $\pm$ 0,204	3,87 $\pm$ 0,123 *	5,80 $\pm$ 0,127 */**

Notes: \* - changes are probable in comparison with the index in PHP (P <0,05);  
\*\* - changes are probable when comparing the indices in patients with NASH (P <0,05);

The content of total cholesterol in blood indicated that it increased by 37.4 and 46.7 ( $p < 0.05$ ) compared with PHPs in patients of 1st and 2nd groups (**table**). Changes in the concentration of TG in the form of a significant increase (respectively, 2.2 and 2.0 times ( $p < 0.05$ )) were recorded in the 1st and 2nd groups of patients. That is, the content in TG in the blood in the comorbid flow of NASH with CKD and obesity were significantly lower than in patients with NASH and obesity.

The study of blood concentrations of proatherogenic lipoprotein fractions indicated a number of changes: the concentration of LDL in the patients of the 1st group was 1.5

times higher than the control group ( $p < 0.05$ ), and in patients of the 2nd group LDL increased in 1.7 times ( $p < 0.05$ ) (**table**). It is also necessary to point out that with the increase in the activity of cytolysis, the content of the cholesterol and LDL in the blood in NASH with comorbidity with CKD and obesity - increased, which may be an important prognostic factor in the progression of atherosclerosis in these patients. Concentration in blood of antiatherogenic lipoproteins - HDL in patients of both groups was significantly lower in comparison with control: in patients of the 1st group - in 1,5 times ( $p < 0,05$ ), in 2nd group - 1,7 times ( $p < 0.05$ ). As can be seen from the results of the study, the maximum suppression of

HDL synthesis (Table 1) was observed in patients of the 2nd group, indicating a minimum level of protection of endothelial vessels from free radical aggression and atherogenic fractions of blood lipids. The result of these changes was a significant increase in the index of atherogenicity in patients of both groups of observation: the 1st group - 2.2 times, the 2nd group - 2.0 times with the maximum changes in the index in patients with NASH, CKD and obesity, which testifies on the one hand, the presence of significant risk factors for the progression of atherosclerosis in these patients on the background of obesity, and on the other - on the favorable pathogenetic situation with regard to the progress of NASH. Thus, the development of NASH in patients with CKD and obesity is accompanied by a significant disorder of dis-hyperlipidemia with the highest among groups comparing with the increase in the content of cholesterol and low-density proatherogenic lipoprotein, a possible decrease in high-density anti-atherogenic lipoprotein and an increase in the atherogenicity index.

The results of the study showed that in patients with NASH, a significant increase in the content of NO in the blood was detected in comparison with the index in PHP ( $p < 0,05$ ) (Table 1) in group 1 - in 2,1 times, in the 2nd group - in 2,6 times ( $p < 0,05$ ). The role of nitrosative stress in the pathogenesis of NASH was proved, the confirmation of which is the increase in the concentration of nitrosothiols, peroxynitrite and other metabolites NO in the blood [2, 10]. Increased peroxynitrite formation due to the generation of NO by leukocytes is an important aspect of the damaging effect and inflammation process in NASH [3]. Pathological hyperproduction of NO by endothelial cells and leukocytes from inflammatory infiltrates in the liver contributes to the development of nitrosative stress in NASH. The established hypernitrate in blood may also be considered compensatory in response to hyperproduction of ET-1 in all observational groups. Thus, the content of ET-1 exceeded the index in PHP, respectively, in patients in the 1st group in 1.7 times, in the 2nd group - in 3.0 times ( $p_{1-2} < 0,05$ ). Confirmation of the presence of endothelial dysfunction (ED) in patients with NASH with CKD resulted in a probable growth of the number of desquamated endothelial cells (DEC) in the 2nd group of patients in 1.9 times ( $p_2 < 0,05$ ). Generation by neutrophils during the exacerbation of NASH of a significant number of active forms of oxygen and nitrogen and hyperproduction of endothelial cells and endometrial lymphocytes with progressive damage to the endothelium (growth of DEC) leads to significant ED, accompanied by mosaic angiospasm of the arteries due to hyperproduction of ET-1 and parietic vasodilatation of the veins of the portal vein system because of the hyperproduction of NO.

**Conclusions** Thus, in patients with non-alcoholic steatohepatitis on the background of obesity lipid distress syndrome with an increase in total cholesterol in blood, low density proatherogenic lipoproteins, and a deficiency in anti-atherogenic high-density lipoproteins is characterized. In a comorbid flow of non-alcoholic steatohepatitis and chronic kidney disease stage I

and II on the background of obesity a deeper lipid imbalance (hypertriacylglycerolemia (2.1 times,  $p < 0,05$ ), hypercholesterolemia (1.5 times,  $p < 0,05$ ), including in the low density lipoprotein (1.8 times,  $p < 0,05$ ), decrease in the content of high density lipoprotein (1.8 times,  $p < 0,05$ ), increase in the atherogenic index (at 2, 7 times,  $p < 0,05$ ), as well as hyperproduction of the endothelium of the NO and lymphocytes with progressive damage to the endothelium (growth of DEC)) was confirmed.

**The prospect of further scientific research in this direction** is the development of a method for the early prevention of non-alcoholic steatohepatitis on the background of obesity and the accompanying CKD of the 1st and 2nd stage.

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