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INFLUENCE OF TOXIC COMBUSTION PRODUCTS ON THE STATE OF INTESTINAL MICROBIOTA IN THE PATHOGENESIS OF NON-ALCOHOLIC FATTY LIVER DISEASE IN FIREFIGHTERS

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Abstract. Medical and psychological support of professional activity of firefighters does not take into account their chronic poisoning by toxic products of burning. Such diagnoses are made to the firefighters of the State Fire Service of the Federal Fire Service of EMERCOM of Russia only in case of acute poisoning. Insufficient attention paid to the impact of fire-specific toxicants, including dioxins, does not allow timely detection of disturbances in the regulation of physiological systems of firefighters' organisms, which leads to the emergence of diseases.
The aim of the study was to determine the effect of dioxins on the state of the microbiota.
Materials and research methods. The study involved 246 male patients, of whom 121 were employees of the State Fire Service of the Federal Fire Service of EMERCOM of Russia and 125 were civilians and rescuers of EMERCOM of Russia.
Research results and their analysis. The data obtained in the study of the blood microbiota by chromatography-mass spectrometry of microbial markers indicate a pronounced intestinal dysbiosis in firefighters with non-alcoholic fatty liver disease due to decreased number of microbial markers of the normal microbiota along with increased total number of microbial markers and opportunistic microorganisms.
The dysbacteriosis revealed in the firefighters having no health complaints can be a predictor of liver pathology development that testifies to necessity of control and timely correction of microecological status.

Key words: *combustion products, dioxins, dysbiosis, Firefighterstoxic microbiota.*

Conflict of interest. The authors declare no conflict of interest

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ВЛИЯНИЕ ТОКСИЧНЫХ ПРОДУКТОВ ГОРЕНИЯ НА СОСТОЯНИЕ МИКРОБИОТЫ КИШЕЧНИКА У ПОЖАРНЫХ В ПАТОГЕНЕЗЕ НЕАЛКОГОЛЬНОЙ ЖИРОВОЙ БОЛЕЗНИ ПЕЧЕНИ

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Резюме. Медико-психологические сопровождение профессиональной деятельности пожарных не учитывает их хронического отравления токсичными продуктами горения. Такие диагнозы сотрудникам Государственной противопожарной службы Федеральной противопожарной службы (ГПС ФПС) МЧС России ставятся только при острых отравлениях. Недостаточное внимание, уделяемое воздействию токсикантов, специфических для пожаров, в том числе диоксинов, не позволяет своевременно выявлять нарушения регуляции физиологических систем организма пожарных, что приводит к возникновению болезней.
Цель исследования – определить влияние диоксинов на состояние микробиоты.
Материалы и методы исследования. В исследовании участвовали 246 пациентов – мужчин, из них 121 – сотрудники ГПС ФПС МЧС России; 125 – гражданские лица и спасатели МЧС России.

Результаты исследования и их анализ. Результаты исследования микробиоты крови методом хромато-масс-спектрометрии микробных маркеров свидетельствуют о выраженном дисбиозе кишечника у пожарных, страдающих неалкогольной жировой болезнью печени (НЖБП), обусловленном снижением количества микробных маркеров нормальной микробиоты, на фоне увеличения общего количества микробных маркеров и условно-патогенной флоры. Дисбактериоз, выявленный у пожарных, не предъявляющих жалоб на состояние здоровья, может являться предиктором развития патологии печени, что свидетельствует о необходимости контроля и своевременной коррекции микроэкологического статуса.
Ключевые слова: *диоксины, дисбактериоз, микробиота кишечника, неалкогольная жировая болезнь печени, пожарные, токсичные продукты горения*

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Introduction

The professional activity of firefighters in conditions of extreme influence of physical, chemical and psychophysiological factors of fires is one of the most dangerous kinds of activity.

The urgency of revealing the chronic intoxication by substances with cumulative effect is determined by the lack of registration in the system of Russian Ministry of Emergency Situations the duration of the influence of toxic products of burning on the organism of firemen. Substances with a high cumulative effect and a very long elimination period include persistent organic pollutants, among which there are dioxins and dioxin-like substances [1]. Analysis of the morbidity of firefighters, presented by the scientific department "Medical registry of the Ministry of Emergency Situations of Russia" of the All-Russian Center of Emergency and Radiation Medicine named after A.M. Nikiforov of EMERCOM of Russia, testifies that the structure of morbidity of firefighters differs from the average values in the population.

Thus, in contrast to population data, where cardiovascular diseases prevail, firefighters face mostly diseases of the gastrointestinal tract (GIT), a quarter of which accounts for non-alcoholic fatty liver disease (NAFLD).

Liver damage, manifested by its fatty degeneration, is one of the main manifestations of intoxication and is accompanied, as a rule, by disorders of fat-soluble vitamin metabolism, porphyrin metabolism and insulin regulation [2].

Despite the fact that dioxins (even in extremely low concentrations) have a negative impact on metabolic processes, mechanisms of intoxication in firefighters are poorly studied. In chronic poisoning by dioxins there is a mutual aggravation of interaction between intestinal microbiota and immune system[3-5].

Y.I. Chernyak noted that cellular enzymatic disorders associated with the activation of detoxification enzymes of such xenobiotics as cytochrome CYP1A2 through aryl hydrocarbon receptor complex with dioxins can lead to disruption of hepatocyte metabolism [6]. One of the mechanisms of gut microbiota composition disorders are changes in the process of synthesis and excretion of bile components, which lead to disorders of internal organ function and microbiota composition. Currently, increased interest in the role of the microbiota in the formation of various pathologies is due to the widespread introduction of innovative molecular genetic DNA sequencing technologies that allow to identify numerous bacterial species that cannot be cultivated [7].

The aim of the study was to investigate the complex effect of persistent organic pollutants, component of toxic products of combustion, on the organism of firefighters during firefighting; to analyze the peculiarities of liver diseases formation in persons of this contingent.

Materials and methods of the study. The study covered 246 male patients, including 121 state fire-fighting service personnel of the Federal Fire-Fighting Service of the Russian Ministry of Emergency Situations and 125 civilian rescuers of the Russian Ministry of Emergency Situations (hereafter, "civilians"). The age of the subjects was 31-46 years; mean age was (38.5±7.5) years. All employees of the state fire-fighting service of the Russian Ministry of Emergency Situations had at least 5 years of work experience in firefighting. None of the civilians were exposed to toxic products of combustion. The groups of Russian EMERCOM personnel and civilians were divided into persons suffering from nonalcoholic fatty liver disease (128), including 63 EMERCOM of Russia and 65 persons of other professions, and those without NAFL (118), including 58 EMERCOM of Russia and 60 persons of other professions.

Based on the blood lipid dioxin concentration estimation the firefighters were divided into 3 groups: Group 1 consisted of 41 people with blood lipid dioxin concentration under 100 pg/g; Group 2 consisted of 37 people with lipid concentration from 101 to 350 pg/g; Group 3 consisted of 43 people with lipid concentration over 350 pg/g. The control group consisted of 125 people with blood lipid dioxin concentrations below 55 pg/g lipids. In 36% of the observed firefighters, blood lipid dioxin concentrations exceeded 350 pg/g lipids. Twenty-five percent of firefighters with NAFL had dioxin concentrations above 350 pg/g lipids; 14% had 101 to 350 pg/g lipids; and 11% of firefighters with NAFL had less than 100 pg/g lipids.

For reference: pg (picogram) is 10-12 grams, i.e., 350 pg of dioxins in 1 g of lipids

Dysbiosis, diagnosed in accordance with OST 91500.11.0004-2003, was determined in 104 firefighters (86%), of which 52% were firefighters with a concentration

of dioxins above 350 pg/g lipids; 21% — firefighters with a concentration from 101 to 350 pg/g lipids; 13% — firefighters with a concentration of less than 100 pg/g lipids.

Exclusion criteria were: cancer, gross somatic pathology, and alcohol abuse.

To assess the state of intestinal microbiota we performed gas chromatography-mass spectrometry (GC-MS) study of microbial markers in blood [8]. The method is based on quantitative determination of microbial markers (fatty acids, aldehydes, alcohols and sterols) directly in clinical material. This method enables the decomposition of the entire pool of microbial markers, which allows the contribution of each of the hundreds of species of microorganisms inhabiting different systems and organs to be assessed.

Whole venous blood with heparin or ethylenediaminetetraacetic acid (EDTA) was pipetted into a 1.5 ml vial with a screw cap and Teflon-lined gasket and dried in the thermostat at 80 °C with 40 µl methanol added to accelerate drying, while the cap was removed. To the thickened sample, 400 µl of 1M hydrochloric acid (this means that 1 L of solution contains 1 mol HCl = 36.5 g HCl) in methanol was added, the lid was tightly screwed on, and acid methanolysis was performed at 80 °C for one hour. To the cooled reaction medium 300 ng (ng — nanograms — 10-9 grams) of the standard, tridecanoic acid deuteromethyl ester dissolved in hexane, was added. The mixture was then extracted in two 200 µl portions of hexane by shaking it on a vortex and allowing it to stand for 5 min at room temperature. The combined extract was transferred to a clean vial, dried for 5-7 min at 80°C, and the dry residue was treated with 20 l of N, O-bis(trimethylsilyl)-trifluoroacetamide for 15 min at 80°C with the lid closed. To the reaction mixture 80 µl of hexane was added — in this form the sample is suitable for analysis for one week if it is hermetically sealed and no evaporation occurs.

For the analysis, a 2 µl mixture of esters was injected into the injector of an Agilent 7890 gas chromatograph with an Agilent 5975C mass-selective detector (Agilent Technologies, USA) through an automatic sample introduction system (autosampler), which provides reproducibility of the retention times of chromatographic peaks and increases the accuracy of automatic data processing. The chromatographic separation of the samples was carried out on a capillary column with methyl silicone grafted phase HP-5ms (Agilent Technologies, USA), the length of 25 m and an inner diameter of 0.25 mm; the carrier gas was helium. Analysis mode — programmed, column thermostat heating rate 7 °C/min in the range 135-320 °C. Delay time at initial temperature — 1.5 min. Evaporator temperature — 250 °C, interface — 250-300 °C.

Results of the study and their analysis. The results of the WHO-TEQ estimation of the blood serum dioxin concentration in lipids of the Federal Fire-fighting Service of Russia and the control group examined show a 16-fold increase in the blood lipid dioxin concentration in firemen compared to the control group.

The analysis of correlation between concentration of blood lipid dioxins and level of microbiota according to OST 91500.11.0004-2003 shows the reduction of concentration of normal microflora of bifidobacteria, eubacteria and lactobacilli among firemen. The decrease in normal microflora values depended on the content of blood lipid dioxins. A more pronounced dysbacteriosis was observed in the groups with a higher concentration of blood lipid dioxins. Along with a decrease in the normal flora there was an increase in the conditionally pathogenic microflora Bacteroides fragilis, Fusobacterium/ Haemophylus, Clostridium perfringens, Peptostreptococcus anaerobius, Enterobacteriacae (E. coli), Staphylococcus intermedius, Bacillus cereus, Bacillus megaterium, Mycobacterium/Candida compared to firefighters with lower blood lipid dioxin concentration as well as control group.

Analysis of the quantitative content in the blood of microbial markers of different groups of microorganisms showed a number of significant differences in the groups examined. A significant increase in the number of microbial markers of aerobic or facultative gram-positive cocci was observed in firefighters with NAFL compared to patients with NAFL of other professions and healthy firefighters. There was a significant increase in microbial markers of aerobic or facultative gram-positive cocci in healthy firefighters compared with healthy individuals in other professions, and a number of indicators exceeded the limits of reference values (Table 1).

The data presented in Table 2 indicate that the increase in the number of microbial markers of opportunistic pathogenic flora in firefighters is mainly due to anaerobic microorganisms. When examining the microbiota in the blood (adherent microbiota) in all examined firefighters was found to increase by more than 35% the total number of microbial markers compared with the examined persons of other professions. At the same time the number of conditionally pathogenic microflora increased by more than 2 times. Also according to a number of indicators there was

Таблица 1 /Table No.1

Содержание микробных маркеров аэробных или факультативных Гр (+) кокков у обследованных в группах наблюдения, M±m, кл/г ×10^5

Assessment of the content of microbial markers of aerobic or facultative Gr (+) cocci in the observation groups, M±m, cells/g ×10^5

Показатель Indicator	Больные НЖБП Patients with nonalcoholic fatty liver disease, n=128		Не страдающие болезнями ЖКТ Not suffering from gastrointestinal diseases, n=118	
	пожарные / firefighters, n=63	контроль / control, n=65	пожарные / firefighters, n=58	контроль / control, n=60
Streptococcus (оральные /oral)	362,4±6,8*#	224,5±5,1#	141,5±2,7*	57,1±1,4
Staphylococcus intermedius	4223,7±32,7*#	2682,7±27,4#	527,3±4,6*	542,4±8,5
Enterococcus	312,4±4,8*#	214,6±1,3#	72,7±2,5*	51,7±1,2
Streptococcus mutans	5218,3±21,5*#	2048,2±23,8#	1432,6±22,4*	198,9±3,1

* различия достоверны (p<0,05) по сравнению с аналогичной контрольной группой (КГ); # различия достоверны (p<0,05) по сравнению с группой здоровых пожарных

* differences are significant (p<0.05) compared to the same control group (CG); # differences are significant (p<0.05) compared to the healthy firefighters group

a reliable increase in conditionally pathogenic microflora in firefighters with NAFL relative to patients of other professions. It is worth noting a reliable decrease in firefighters in comparison with patients with NAFL and healthy other professions, as well as a clear trend to a decrease in microbial markers (Bifidobacterium) in healthy firefighters relative to healthy other professions. The multidirectional changes in the microbiota, a decrease in bifidobacteria, actinomycetes and, at the same time, an increase in bacteroides and ruminococci in firefighters with NAFL in comparison with NAFL patients from the control group, draw attention.

The results of the evaluation of the content of microbial markers of aerobic or facultative Gr(+) bacilli indicate that firefighters suffering from NAFL have significantly higher values of microbial markers of aerobic or facultative Gr(+) bacilli compared with patients from the control group and healthy firefighters. Noteworthy is the significant decrease (Lactobacillus) in all firefighter groups compared to other occupational groups (Table 3).

Evaluation of microbial markers of aerobic or facultative Gr(-) bacilli showed a significant increase in markers of some microorganisms (Helicobacter pylori, h 18, Enterobacteriacae (E. coli)) in firefighters with NAFL compared to those in other occupations and increased markers of some microorganisms (Helicobacter pylori, h 18, Achromobacter, Enterobacteriacae (E. coli)) compared to sick and healthy control groups (Table 4).

The results of the analysis of the content of microbial markers of fungi, viruses and other microorganisms in firefighters with NAFL indicate significantly higher content indicators (Mycobacterium/ Candida, Streptomyces, Micr. fungi, sitosterol) than in patients from the control group. At the same time, the content of microbial markers in firefighters with NAFL is also significantly higher than in healthy firefighters. At the same time the indices of content of microbial markers in healthy firefighters are reliably higher than in the patients and healthy control group.

The increased viral load (Herpes) in firefighters of both groups also draws attention (Table 5).

Table 6 shows the distribution of reduced content of normal microbiota, draws attention to the decrease in the number of microbial markers Bifidobacterium, Eubacterium/Cl. Coocoides and Lactobacillus in the blood of firefighters both with and without GI disease.

It was found that more than half of the firefighters with NAFL had increased total number of microbial markers in the blood. At the same time, the main changes in the microbiota composition occur due to the increase of

Таблица 2 /Table No.2

Содержание микробных маркеров анаэробных микроорганизмов у обследованных в группах наблюдения, M±m, кл/г ×10^5

Assessment of the content of microbial markers of anaerobic microorganisms in the observation groups, M±m, cells/g ×10^5

Показатель Indicator	Больные НЖБП Patients with nonalcoholic fatty liver disease, n=128		Не страдающие болезнями ЖКТ Not suffering from gastrointestinal diseases, n=118	
	пожарные / firefighters, n=63	контроль / control, n=65	пожарные / firefighters, n=58	контроль / control, n=60
Actinomyces viscosus	2132,6±18,4*#	1844,5±23,5*	178,5±2,3*	89,4±1,5
Bacteroides fragilis	836,4±5,4*#	72,1±1,4*	541,3±1,3*	112,4±1,6
Bacteroides hypermegas	248,7±4,2*#	42,5±1,1*	172,3±2,7*	36,5±1,4
Bifidobacterium	1489,2±21,5*#	4631,3±18,3*	1978,2±23,6*	6243,4±19,5
Butyrivibrio/ Cl/ fimetarum	3142,4±12,3*	2437,2±16,4*	1024,6±9,6	-
Cl. Difficile	689,4±3,7#	492,2±7,6*	298,7±5,4	128,7±6,5
Clostridium hystoliticum	862,5±14,3*#	82,3±2,3*	378,4±13,4*	-
Clostridium perfringens	752,2±14,3*#	579,1±9,3*	142,4±3,2*	24,7±2,1
Clostridium ramosum	4126,3±37,2*#	2372,4±23,7*	3825,3±27,3*	841,4±5,7
Eubacterium	26,7±1,1*#	72,3±2,5*	28,3±1,4*	79,5±2,4
Eubacterium lentum (группа A)	524,1±6,3*#	292,2±5,6*	334,5±4,7*	128,5±3,1
Eubacterium moniliforme sbsp	6542,5 ± 58,6	3218,5 ± 37,4	0,0	0,0
Eubacterium/Cl. Coocoides	3824,3±34,6	5916,7±53,7	4482,0±23,6	7214,4±31,5
Fusobacterium/ Haemophylus	187,4±3,7*#	45,2±1,4	152,8±2,4	24,1±1,2
Peptostreptococcus anaerobius (Гр. 1)	756,1±11,4#	256,0±6,5	421,5±4,1	0,0
Porphyromonas	189,4±3,8*#	525,7±4,3	72,1±2,4	36,8±1,4
Prevotella	24,1±0,7	82,3±2,1	19,3±1,6	32,6±0,9
Propionibacterium	1734,2±4,7**	293,6±8,2	1245,1±7,3*	97,12±4,1
Propionibacterium acnes	182,3±2,7*#	0,0	214,5±3,2	0,0
Propionibacterium jensenii	952,4±5,4	398,7±3,6	724,3±2,4*	218,4±2,3
Propionibacterium spp. (P. freuden)	986,4±2,4*#	4125,3±18,6	1746,2±6,5	4536,8±28,4
Ruminicoccus	1745,2±12,3*#	426,3±14,5	1264,5±13,4	352,4±11,6
Актиномицеты / Actinomycetes	286,5±2,7*	1234,6±4,1	318,4±2,6*	1190 ± 21,3

*различия достоверны (p<0,05) по сравнению с аналогичной КГ; #различия достоверны (p<0,05) по сравнению с группой здоровых пожарных

* differences are significant (p<0.05) compared to the same control group (CG);

differences are significant (p<0.05) compared to the healthy firefighters group

Таблица 3 /Table No.3

Содержание микробных маркеров аэробных или факультативных Гр(+) палочек, M±m, кл/г ×10^5

Assessment of the content of microbial markers of aerobic or facultative Gr (+) bacilli, M±m, cells/g ×10^5

Показатель Indicator	Больные НЖБП Patients with nonalcoholic fatty liver disease, n=128		Не страдающие болезнями ЖКТ Not suffering from gastrointestinal diseases, n=118	
	пожарные / firefighters, n=63	контроль / control, n=65	пожарные / firefighters, n=58	контроль / control, n=60
Bacillus cereus	252,2±2,5*#	182,4±6,5	156,7±4,9	21,8±1,2
Nocardia (14:1d11)	252,9 ± 7,5* #	281,1±7,3	196,2±6,7	263,4±3,1
Nocardia asteroides	326,8±2,9*	482,6±4,3	512,8±5,6	468,5±8,2
Lactobacillus	3225,3±8,6*	5283,3±6,2	3615,4±7,4	6834,4±3,6
Rhodococcus	527,4±134,8*	286,5±8,2	295,4±6,3	482,3±3,7
Bacillus megaterium	5236,5±42,9*#	3982,4±23,4	4621,6±34,1	2380,4±27,4

*различия достоверны (p<0,05) по сравнению с аналогичной КГ; #различия достоверны (p<0,05) по сравнению с группой здоровых пожарных
* differences are significant (p<0.05) compared to the same control group (CG);
# differences are significant (p<0.05) compared to the healthy firefighters group

Таблица 4 //Table No.4

Содержание микробных маркеров аэробных или факультативных Гр (-) палочек, M±m, кл/г ×10^5

Assessment of the content of microbial markers of aerobic or facultative Gr (-) bacilli, M±m, cells/g ×10^5

Показатель Indicator	Больные НЖБП Patients with nonalcoholic fatty liver disease, n=128		Не страдающие болезнями ЖКТ Not suffering from gastrointestinal diseases, n=118	
	пожарные / firefighters, n=63	контроль / control, n=65	пожарные / firefighters, n=58	контроль / control, n=60
Achromobacter	156,4±2,4##	162,5±1,7	148,2±1,2	52,3±1,7
Campylobacter mucosalis	232,4±3,2	126,3±1,4	57,5±1,1	44,8±1,6
Helicobacter pylori, h 18	176,4±2,5*#	26,2±2,7	124,8±3,5*	17,5±0,9
Сем. Enterobacteriacae (E. coli)	59,7±1,3*##	0	52,7±1,5*	0

*различия достоверны (p<0,05) по сравнению с аналогичной КГ; #различия достоверны (p<0,05) по сравнению с группой здоровых пожарных
* differences are significant (p<0.05) compared to the same control group (CG);
# differences are significant (p<0.05) compared to the healthy firefighters group

opportunistic-pathogenic flora against the background of a decrease in the normal microflora.

The vast majority of firefighters with NAFL have increased concentrations of Bacteroides fragilis, Fusobacterium/ Haemophylus, Clostridium perfringens, Peptostreptococcus anaerobius, Enterobacteriacae (E. coli), Staphylococcus intermedius, Bacillus cereus, Bacillus megaterium, Mycobacterium/ Candida.

Conclusion

The research of the blood microbiota by chromatography-mass-spectrometry method of microbial markers testifies to the presence of the expressed intestinal dysbiosis at the examined firemen which is much more expressed at the firemen with NAFL which is caused by the increase of the total number of microbial markers and conditionally-pathogenic flora as well as the reduction of the number of microbial markers of normal microbiota.

It is known that, in chronic liver disease, the concentration of normal microflora, which includes bifidobacteria, bacteroides and lactobacilli, decreases. At the same time, the concentration of conditionally pathogenic flora increases. Nonalcoholic fatty liver disease is accompanied by

Таблица 5 /Table No.5

Содержание микробных маркеров грибов, вирусов и прочих микроорганизмов в группах наблюдения, M±m, кл/г ×10^5

Assessment of the content of microbial markers of fungi, viruses, and other microorganisms in the observation groups, M±m, cells/g ×10^5

Показатель Indicator	Больные НЖБП Patients with nonalcoholic fatty liver disease, n=128		Не страдающие болезнями ЖКТ Not suffering from gastrointestinal diseases, n=118	
	пожарные / firefighters, n=63	контроль / control, n=65	пожарные / firefighters, n=58	контроль / control, n=60
Микр. грибы, кампестерол / Microbial fungi, campesterol	1562,3±23,6*	464,3±25,4	1254,6 ±31,8	637,2±27,5
Микр. грибы, ситостерол / Microbial fungi, sitosterol	764,4±26,5*	242,4±19,7#	518,6±23,7	184,3±9,2
Mycobacterium/ Candida	264,5±3,5*	0,00##	182,5±4,2	0,00
Streptomyces	1825,8±32,6*	256,4±14,3#	1687,3±41,5*	189,6±12,4
Herpes	582,6±12,4*	282,3±8,9	497,7±14,7	126,4±7,6
Pseudonocardia	512,57±8,9*	97,5±7,4	482,6±12,5	72,9±3,2

*различия достоверны (p<0,05) по сравнению с аналогичной КГ; #различия достоверны (p<0,05) по сравнению с группой здоровых пожарных
* differences are significant (p<0.05) compared to the same control group (CG);
# differences are significant (p<0.05) compared to the healthy firefighters group

Таблица 6 /Table No.6

Распространенность изменения содержания микробных маркеров микрофлоры в группах наблюдения (ОСТ 91500.11.0004-2003), %

Prevalence of changes in the content of microbial markers of microflora in observation groups (OST 91500.11.0004-2003), %

Показатель Indicator	Больные НЖБП Patients with nonalcoholic fatty liver disease, n=128		Не страдающие болезнями ЖКТ Not suffering from gastrointestinal diseases, n=118	
	пожарные firefighters, n=63	контроль control, n=65	пожарные firefighters, n=58	контроль control, n=60
Сниженное содержание микробных маркеров нормальной микрофлоры в крови Reduced content of microbial markers of normal microflora in blood				
Bifidobacterium (5067)	62,0	12,0	46,0	0,0
Lactobacillus (6613)	58,0	18,0	43,0	0,0
Eubacterium/Cl. Coocoides (6912)	48,0	12,0	24,0	0,0
Eubacterium (59)	51,0	0,0	47,0	0,0
Повышенное содержание микробных маркеров условно-патогенных микрофлоры в крови Increased content of microbial markers of opportunistic microflora in blood				
Bacteroides fragilis	32,0	0,0	21,0	0,0
Fusobacterium/ Haemophylus	23,0	6,0	12,0	2,0
Clostridium perfringens	57,0	41,0	32,0	1,0
Eubacterium	0,0	5,0	4,0	6,0
Peptostreptococcus anaerobius (Гр.1)	72,0	36,0	63,0	0,0
Enterobacteriacae (E. coli)	26,0	0,0	21,0	0,0
Enterococcus (290)	7,0	0,0	0,0	0,0
Staphylococcus intermedius (756)	72,0	48,0	0,0	0,0
Bacillus cereus (23)	67,0	52,0	36,0	0,0
Bacillus megaterium	81,0	32,0	29,0	3,0
Mycobacterium/ Candida	72,0	0,0	49,0	0,0

changes in composition of human microflora, which is manifested by deficiency of obligate microorganisms and microbial insemination of small intestine, resulting in formation of vicious circle, which supports mutual aggravation of pathological changes of intestine and liver. A key link in pathogenesis of formation of intestinal microbiota composition disorders are changes in synthesis and excretion of bile components, which result in dysfunction of internal organs. At the same time excessive bacterial growth leads to increased intake of products of bacterial life activity and putrefaction into the blood stream, which results in disturbances of immune system regulation, increased cytokine production, dysregulation of secretory immunoglobulins (IgA), decreased phagocytic activity, changes of water and electrolyte balance and acid-base equilibrium. Studies have shown the ability of microorganisms to recombinant DNA changes during their phagocytosis, which can lead to the synthesis of specific antibodies to these DNA fragments.

Most of the mechanisms of NAFL are realized through an increase in fat deposition and development of insulin resistance (IR), which has a significant impact on the regulation of metabolism in general. Also a number of products of bacterial life activity have hepatotoxicity. In the pathogenesis of NAFL there is a theory of "two blows" — entry of unesterified fatty acids into the hepatocyte and damage of hepatocytes by oxidative stress. When analyzing the pathogenesis of NAFL, it is necessary to pay special attention to the weighty contribution to the formation of pathology of bile acids dysregulation, which play an important role in lipid metabolism. Activation of systemic inflammatory response against oxidative stress is aggravated by activation of proinflammatory cytokine synthesis by tissue macrophages influenced by increased concentration of bacterial lipopolysaccharides.

The results of the present study indicate that firefighters are characterized by peculiarities in the pathogenesis of NAFL that do not fit into the mechanisms presented in the scientific literature. It should be taken into account that firefighters are exposed to many toxic products of combustion while performing professional tasks. One such product is dioxins, the collective name for polychlorinated biphenyls, which include a large number of different congeners. Despite the fact that the main route of dioxin intake into the human body is alimentary, their commulative properties, high concentration and long exposure to firefighting allow dioxins to accumulate in firefighters' bodies. According to scientific publications, the metabolic activity of dioxins is mediated through the aryl hydrocarbon receptor (AhR), which is a metabolic regulator and is expressed on almost all cells in the body. Through aryl receptors, dioxin affects the regulation of glucose and lipid metabolism and modifies the expression of genes regulating insulin transport and receptor interactions in human adipose tissue, and also affects cholesterol biosynthesis, fatty acid synthesis, adipocyte differentiation and leptin regulation.

Thus, firefighters had a syndrome of excessive growth of intestinal microbiota due to an increase in the number of conditionally pathogenic flora on the background of a decrease in the number of normal microflora. The revealed increases and changes in the composition of the intestinal microbiota suggest the presence of dysbiosis of the intestinal wall flora in the examined firefighters, which seems to be associated with a decrease in both general and local immunity against the background of exposure to toxic products of combustion, including dioxins, when performing professional tasks of firefighting. The presence of severe dysbiosis, typical for firefighters with NAFL, significantly complicates the course of the disease. Manifestations of dysbiosis detected in healthy firefighters can serve as predictors of liver pathology development, which indicates the need for monitoring and targeted correction of somatic condition and microecological status of this contingent.

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