Медицинская иммунология
2023, Т. 25, № 5,
стр. 1205-1212

Краткие сообщения
Short communications

Medical Immunology (Russia)/
Meditsinskaya Immunologiya
2023, Vol. 25, №5, pp. 1205-1212

АКТИВНОСТЬ ЯДЕРНОГО ФАКТОРА ТРАНСКРИПЦИИ κB (NF-κB) В ПОПУЛЯЦИЯХ ЛИМФОЦИТОВ У ДЕТЕЙ С БОЛЕЗНЬЮ ВИЛЬСОНА–КОНОВАЛОВА

Курбатова О.В.[1], Петричук С.В.[1], Купцова Д.Г.[1], Мовсисян Г.Б.[1], Радыгина Т.В.[1], Комарова А.Д.[1], Анушенко А.О.[1], Фрейдлин Е.В.[1], Семикина Е.Л.[1, 2], Потапов А.С.[1, 2], Фисенко А.П.[1]

[1] ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ, Москва, Россия
[2] ФГАОУ ВО «Первый Московский государственный медицинский университет имени И.М. Сеченова» Министерства здравоохранения РФ (Сеченовский университет), Москва, Россия

Резюме. Болезнь Вильсона (БВ) – редкое наследственное заболевание, вызванное дефицитом транспортера АТФ7В. Накопление меди может вызывать повреждение органов и клеток, преимущественно печени. Воздействие меди может модулировать синтез цитокинов посредством молекулярных и клеточных сигнальных путей, включая путь ядерного фактора транскрипции NF-κB. NF-κB является главным регулятором воспаления и гибели клеток, действует как центральное звено между повреждением печени, фиброзом и гепатоцеллюлярной карциномы. Избыток NF-κB-зависимого цитокинового ответа стимулирует воспалительные реакции, но и чрезмерное ингибирование NF-κB может негативно влиять на жизнеспособность гепатоцитов. Метод проточной цитометрии с визуализацией – Amnis ImageStream^X позволяет оценить активность NF-κB (% активированных клеток, в популяциях клеток). Цель работы – оценить активность NF-κB в популяциях лимфоцитов у детей с болезнью Вильсона–Коновалова. Иммунофенотипирование лимфоцитов и оценку уровня транслокации NF-κB проводили у 52 ребенка с БВ и у 25 детей группы сравнения. Массовую концентрацию меди в суточной моче определяли атомно-абсорбционным методом с помощью спектрометра AAnalyst 800. У детей с БВ содержание клеток с транслокацией NF-κB варьировало от 5 до 90% в зависимости от популяции лимфоцитов, наибольший уровень выявлен в В-клетках и составил 57,5 (37-68) %. Показана достоверная разница в распределениях количества клеток с транслокацией NF-κB в популяциях лимфоцитов между БВ и здоровыми детьми (F-критерий, $p < 0,01$). Для детей с БВ в большинстве случаях характерно снижение активности фактора транскрипции NF-κB в популяциях В-клеток (в 43% случаев), Т-хелперов (48%), Т-цитотоксических (44%) и Th17-лимфоцитов (41%). У детей с БВ

Адрес для переписки:

Курбатова Ольга Владимировна
ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ
119991, Россия, Москва, Ломоносовский пр. 2, стр. 1.
Тел.: 8 (499) 134-13-98.
Факс: 8 (499) 134-70-01.
E-mail: putintseva@mail.ru

Address for correspondence:

Olga V. Kurbatova
National Medical Research Center of Children's Health
2 Lomonosovsky Ave, Bldg 1
Moscow
119991 Russian Federation
Phone: +7 (499) 134-13-98.
Fax: +7 (499) 134-70-01.
E-mail: putintseva@mail.ru

Образец цитирования:

О.В. Курбатова, С.В. Петричук, Д.Г. Купцова, Г.Б. Мовсисян, Т.В. Радыгина, А.Д. Комарова, А.О. Анушенко, Е.В. Фрейдлин, Е.Л. Семикина, А.С. Потапов, А.П. Фисенко «Активность ядерного фактора транскрипции κB (NF-κB) в популяциях лимфоцитов у детей с болезнью Вильсона–Коновалова» // Медицинская иммунология, 2023. Т. 25, № 5. С. 1205-1212.
doi: 10.15789/1563-0625-NTF-2799

For citation:

O.V. Kurbatova, S.V. Petrichuk, D.G. Kuptsova, G.B. Movsisyan, T.V. Radygina, A.D. Komarova, A.O. Anushenko, E.V. Freidlin, E.L. Semikina, A.S. Potapov, A.P. Fisenko "Nuclear transcription factor κB (NF-κB) activity in lymphocyte populations in children with Wilson–Konovalov disease", Medical Immunology (Russia)/Meditsinskaya Immunologiya, 2023, Vol. 25, no. 5, pp. 1205-1212.
doi: 10.15789/1563-0625-NTF-2799

DOI: 10.15789/1563-0625-NTF-2799

концентрация меди варьировала от 9,7 до 2582 мкг/сут, Med = 616 (210-1173). Получена прямая зависимость между содержанием меди в моче и уровнем транслокации NF-κB в В-лимфоцитах, r = 0,34, p = 0,016. Активность фактора NF-κB коррелирует с биохимическими маркерами тяжести поражения печени (АЛТ, АСТ, ГГТ) и с содержанием меди в моче. Исследование сигнального пути NF-κB представляется перспективным для большего понимания патогенетических механизмов формирования процессов воспаления и фиброза печени у детей с БВ.

Ключевые слова: дети, болезнь Вильсона–Коновалова, лимфоциты, Th17, Tc17, проточная цитометрия с визуализацией, NF-κB

NUCLEAR TRANSCRIPTION FACTOR κB (NF-κB) ACTIVITY IN LYMPHOCYTE POPULATIONS IN CHILDREN WITH WILSON–KONOVALOV DISEASE

Kurbatova O.V.[a], Petrichuk S.V.[a], Kuptsova D.G.[a], Movsisyan G.B.[a], Radygina T.V.[a], Komarova A.D.[a], Anushenko A.O.[a], Freidlin E.V.[a], Semikina E.L.[a, b], Potapov A.S.[a, b], Fisenko A.P.[a]

[a] *National Medical Research Center of Children's Health, Moscow, Russian Federation*
[b] *I. Sechenov First Moscow State Medical University (Sechenov University), Moscow, Russian Federation*

Abstract. Wilson's disease (WD) is a rare hereditary disease caused by a deficiency of the ATF7B transporter. The accumulation of copper can cause damage to organs and cells, mainly the liver. Copper exposure can modulate cytokine synthesis through molecular and cellular signaling pathways, including the nuclear transcription factor NF-κB pathway. NF-κB is the main regulator of inflammation and cell death, acts as a central link between liver damage, fibrosis and hepatocellular carcinoma. An excess of NF-κB-dependent cytokine response stimulates inflammatory reactions, but excessive inhibition of NF-κB can negatively affect the viability of hepatocytes. Method of flow cytometry with visualization – Amnis ImageStreamX allows to evaluate the activity of NF-κB (% of activated cells in cell populations). The aim: to evaluate the activity of NF-κB in lymphocyte populations in children with WD disease. Immunophenotyping of lymphocytes and assessment of the level of translocation of NF-κB were performed in 52 children with WD and in 25 children of comparison group. The mass concentration of copper in daily urine was determined by atomic absorption method using the AAnalyst 800 spectrometer. In children with WD, the content of cells with NF-κB translocation varied from 5 to 90% depending on the lymphocyte population; the highest level was detected in B cells – 57.5 (37-68) %. A significant difference in distributions of the number of cells with NF-κB translocation between WD and healthy children was shown (F-criterion, $p < 0.01$). In most cases, children with WD are characterized by a decrease in the activity of NF-κB in populations of B cells (in 43% of cases), T helper cells (48%), T cytotoxic (44%) and Th17 lymphocytes (41%). In children with WD, the concentration of copper varied from 9.7 to 2582 mcg/day, Me = 616 (210-1173). A direct relationship was obtained between the copper content in urine and the level of translocation of NF-κB in B lymphocytes, r = 0.34, p = 0.016. The activity of the NF-κB correlates with biochemical markers of the severity of liver damage (ALT, AST, GGT) and with copper content in urine. The study of the NF-κB signaling pathway seems promising for a better understanding of the pathogenetic mechanisms of the formation of inflammation and liver fibrosis in children with WD.

Keywords: children, Wilson–Konovalov disease, lymphocytes, Th17, Tc17, flow cytometry with visualization, NF-κB

Introduction

Wilson's disease (WD) is a rare hereditary disease caused by a deficiency of the ATP7B transporter. The protein encoded by this gene promotes the incorporation of copper into the copper-containing protein, ceruloplasmin. In WD, copper accumulates primarily in the liver and secondarily in other organs, such as the central nervous system. WD in some patients is asymptomatic, while others develop acute liver failure. Verification of the diagnosis requires a combination of clinical signs and diagnostic tests such as decreased serum ceruloplasmin levels, increased

urinary copper excretion, liver biopsy or genetic testing [3, 6].

Copper (Cu) is an essential micronutrient, however, excessive accumulation of copper can cause damage to organs and cells, catalyze the formation of free radicals and trigger lipid peroxidation. Malondialdehyde formed as a result of lipid peroxidation stimulates collagen synthesis, promoting fibrogenesis. The toxic effect of copper on the liver tissue can manifest itself in the form of fatty degeneration of hepatocytes, hepatitis, fibrosis and cirrhosis of the liver [12]. The toxicological and inflammatory effects of Cu have been investigated in various animal models and cells [2]. It has been shown that excessive exposure to Cu can modulate cytokine synthesis through various molecular and cellular signaling pathways, including the NF-κB nuclear transcription factor pathway, the MAPKs pathway, the JAK-STAT pathway, and NLRP3 pathway [2].

Disturbances in the immune system in patients with WD have been described, progressing with an increase in the stage of liver fibrosis and with an increase in the concentration of copper in daily urine [5].

The transcription factor NF-κB is the main regulator of inflammation and cell death, in the development of hepatocellular damage, liver fibrosis and hepatocellular carcinoma, acts as a central link between liver damage, fibrosis and hepatocellular carcinoma, however, inhibition of NF-κB can not only provide beneficial influence, but also negatively affect the viability of hepatocytes, especially with pronounced inhibition of NF-κB [7]. The p50 NF-κB subunit plays a critical protective role in damaged liver by limiting TNFα expression and inflammatory cell recruitment. In an experimental model that mimics chronic liver disease, NF-κB-mice developed more severe neutrophilic inflammation and fibrosis compared to NF-κB$^+$ mice [9]. However, an excess of NF-κB-dependent cytokine response can stimulate inflammatory responses. Therefore, NF-κB activity should be kept under control to maintain immune balance [8].

The modern method of flow cytometry with visualization – Amnis Image Stream X allows to evaluate the activity of the transcription factor NF-κB (% of activated cells in which NF-κB passes from the cytoplasm to the cell nucleus) in various cell populations [1]. NF-κB activity has not been studied in children with WD.

The aim: to evaluate the activity of NF-κB in the populations of lymphocytes in children with WD.

Materials and methods

We examined 52 children with WD aged 6 to 18 years, Me 13.6 (11.0-16.4). The comparison group consisted of 25 healthy children (HC) without somatic, autoimmune, oncological pathology, comparable in age. Immunophenotyping of major and minor populations of lymphocytes in peripheral blood was performed by flow cytometry (Novocyte, ACEA Biosciences, USA). The level of NF-κB translocation was assessed on pre-isolated peripheral blood mononuclear cells (PBMCs) by flow cytometry with imaging (Amnis ImageStreamX Mk II) using the Amnis NF-κB Translocation Kit (Luminex, USA). We used monoclonal antibodies labeled with fluorochromes: CD19-PE, CD4-PE, CD8-PE, CD(16/56)-PE, CD161-PE, CD3-ECD, CD4-PB (Beckman Coulter, USA). The following cell populations were studied: CD3-CD19$^+$ (B lymphocytes); CD3-CD16$^+$/CD56$^+$ (NK cells); CD3$^+$CD4$^+$ (T helpers); CD3$^+$CD8$^+$ (cytotoxic T lymphocytes); CD3$^+$CD161$^+$CD4$^+$ (Th17 lymphocytes); CD3$^+$CD161$^+$CD8$^+$ (cytotoxic T17 lymphocytes – Tc17).

The mass concentration of copper in daily urine was determined by the atomic absorption method using an AAnalyst 800 spectrometer (N.F. Izmerov Research Institute of Occupational Medicine).

Statistical processing of the obtained data was carried out using the Statistica 10.0, descriptive statistics of the indicators are presented in the form: Me ($Q_{0,25}$-$Q_{0,75}$). To assess the significance of differences between groups, the nonparametric Mann–Whitney test, the Wilcoxon test for conjugated pairs, and the Fisher test were used. Differences were considered statistically significant at $p < 0.05$.

Results and discussion

The content of cells with NF-κB translocation in different populations differed significantly both in the group of children with WD and in the comparison group (Wilcoxon test for conjugated pairs, $p = 0.000$). The highest percentage was characteristic of the B cell population (WD – 57.5 (37-68) %; HC – 58.4 (43-79) %), the smallest for T cells (WD – 16.4 (13-24)) %; HC – 17.6 (16-21) %), intermediate values were found in the NK cell population (WD – 27.1 (19-43) %; HC – 27.8 (20-34) %). The Mann–Whitney test did not reveal significant differences between the groups in terms of the content of cells with NF-κB translocation, however, due to the large scatter of indicators in the group with WD, a significant difference was shown in the distribution of indicators in groups according to the F-criterion (Figure 1A, $p < 0.01$). The distributions of NF-κB activity in the studied populations differ, with the exception of B cells.

In children with WD, NF-κB activity in Tc17 lymphocytes was 20.8% (12.5-32.5) and exceeded NF-κB activity in T helpers ($p = 0.024$), Th17 ($p = 0.000$) and cytotoxic T – lymphocytes ($p = 0.001$) (Figure 1B). In children of the control group, NF-κB activity in Th17 lymphocytes was 19.3% (17-21) and

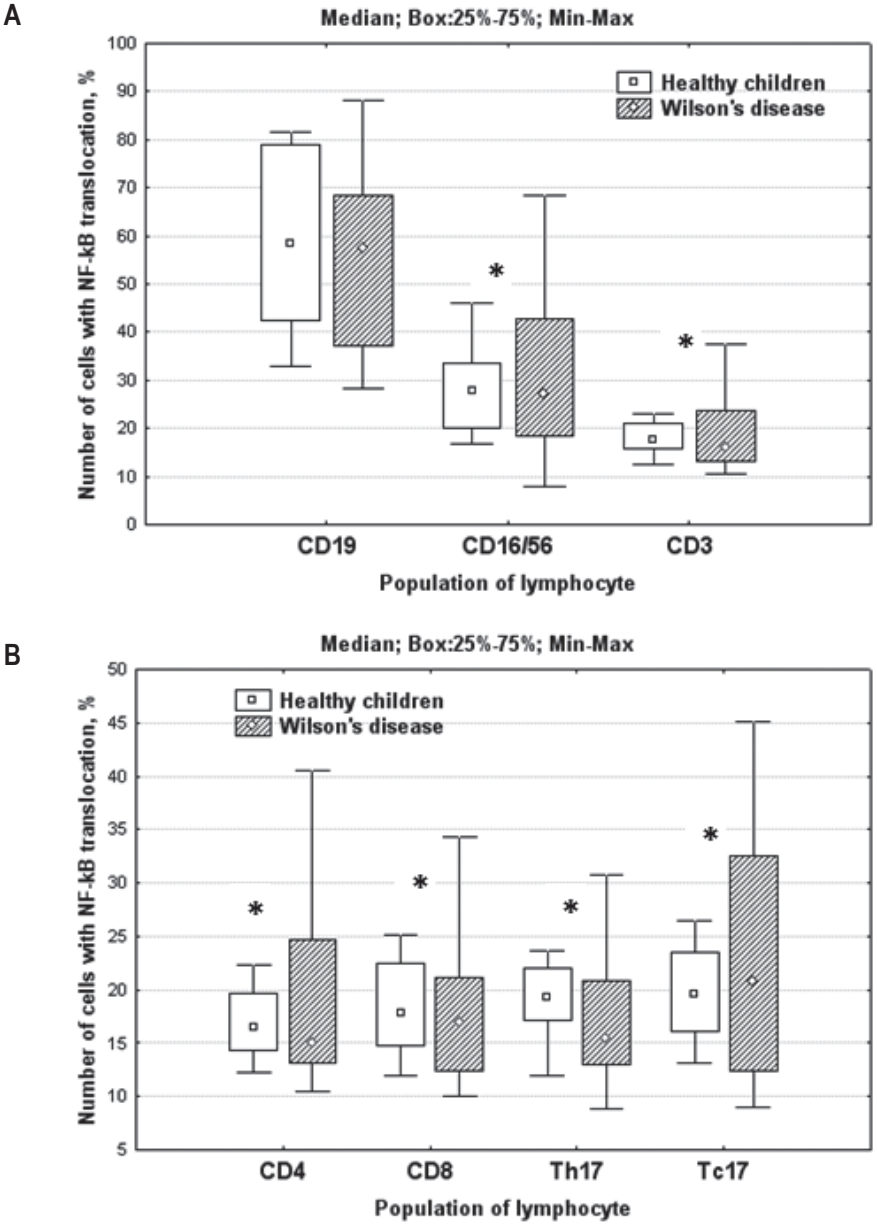

Figure 1. Content of the main and small populations of lymphocytes with NF-κB translocation in children with wd and healthy children

Note. *, F-test, $p < 0.01$.

exceeded NF-κB activity in T helpers ($p = 0.033$) (Figure 1C).

To identify the features of NF-κB activity in the group of children with WD, we performed a frequency analysis of the distribution of indicators in all the studied populations of lymphocytes. It was found that the content of cells with NF-κB translocation in B lymphocytes varied from 25 to 90%. At the same time, only in 45% of patients with WD, the content of cells with NF-κB translocation in B lymphocytes corresponded to the level of the comparison group, in 43% of patients this indicator was reduced, and in 12% of patients it was increased (Figure 2A).

The content of cells with NF-κB translocation into NK cells varied from 5 to 70% of activated cells, only in 29% of patients with WD the content of NF-κB in NK cells corresponded to the level of the control group, in 42% of patients this indicator was higher than the control group, decreased in 29% of patients (Figure 2B). The content of cells with NF-κB translocation in the T helper ($CD4^+$) population was characterized by less variability – from 10 to 50% of activated cells, and in most children with WD (48%) a decrease in this indicator was noted, and corresponded to the level of the comparison group only in 21% of patients (Figure 2C). A similar distribution was also characteristic of the T-cytotoxic lymphocyte

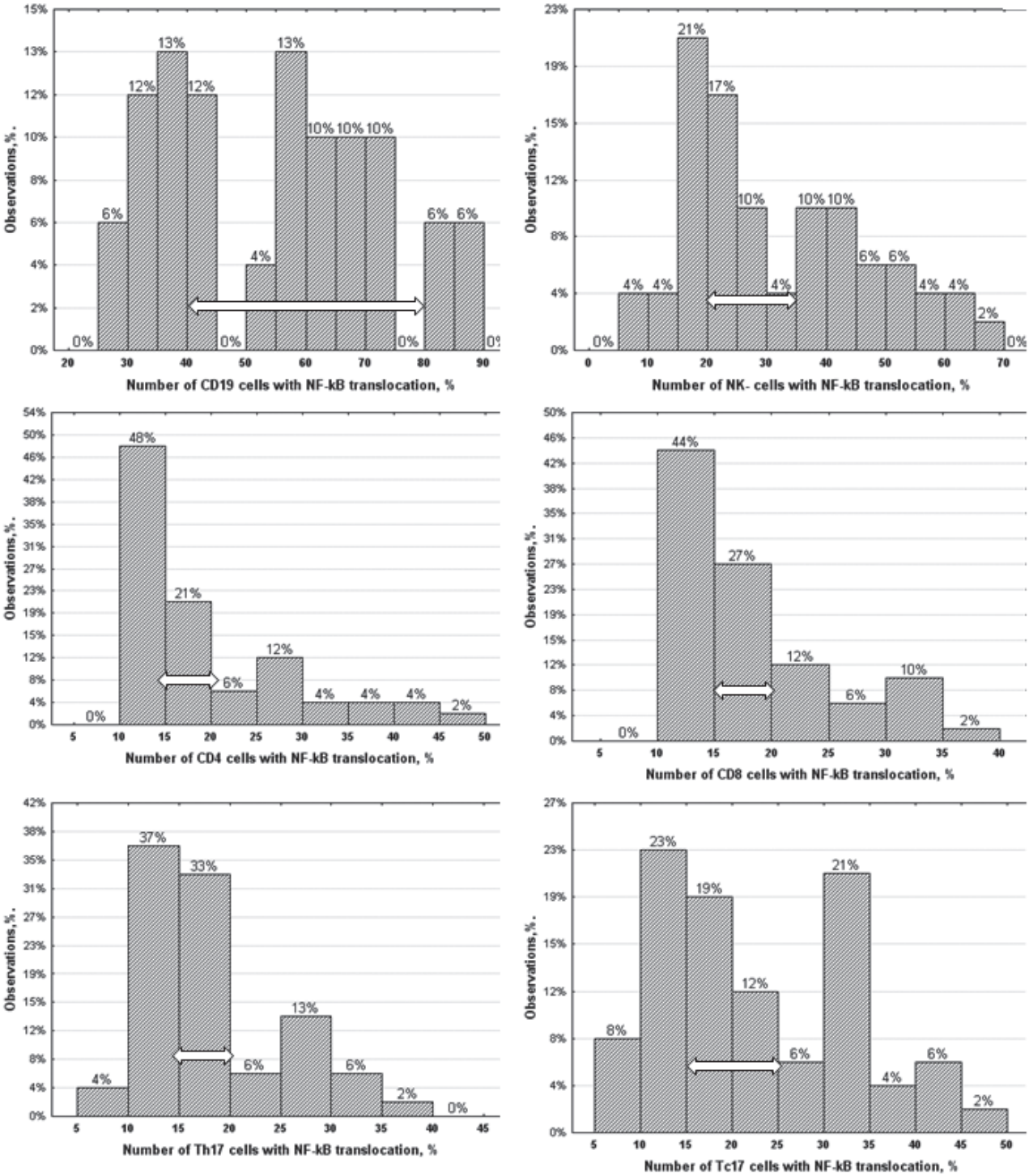

Figure 2. Distribution of the number of cells with NF-κB translocation in different populations of lymphocytes in children with WD

population: a decrease in 44% and an increase in 29% of patients (Figure 2D). The content of cells with NF-κB translocation in the Th17 lymphocyte population was reduced in 41%, increased in 26% of patients (Figure 2E). In the Tc17 lymphocyte population, a decrease was found in 31% and an increase in 38% of patients (Figure 2F). An increase in NF-κB activity was most characteristic of the NK cell population (42% of observations), and the increase in the content of cells with NF-κB translocation in children with WD reached 70%.

It is interesting to note that in children with WD, the translocation level indicators are comparable with those of the comparison group (medians do

not differ significantly), in contrast to children with autoimmune diseases (IBD, psoriasis), who have an increased level of NF-κB activity [4, 10, 11].

The number of lymphocytes with reduced NF-κB activity in the studied populations ranged from 29 to 48% of patients. This fact may be explained by the fact that lymphocytes reflect the intensity of inflammatory processes in the liver, which is consistent with the data of Luedde T. that complete blockade of NF-κB in hepatocytes enhances liver damage [7].

At the next stage of the study, we analyzed the correlations between the content of major and minor populations of lymphocytes and the percentage of activated cells in these populations. An analysis of cellular immunity parameters in this cohort of children with WD confirms the previously published data on the presence of an increase in the content of T helpers, regulatory T cells, Th17 lymphocytes and activated T helpers against the background of a decrease in cytotoxic T lymphocytes and NK cells relative to the comparison group [5].

An inverse correlation was found between the relative number of B cells and the number of B lymphocytes with NF-κB translocation, $r = -0.347$, $p = 0.012$. Correlation analysis between the content of other populations of lymphocytes and activated cells with NF-κB translocation in these populations did not reveal statistically significant relationships. The level of NF-κB translocation in the lymphocyte populations of children with WD also did not depend on age.

Significant sensitive markers of liver damage and the proportion of affected cells with NF-κB translocation were identified. Thus, the concentration of alanine aminotransferase (ALT) increased with an increase in the number of activated cells with NF-κB translocation in the populations of T helpers ($r = 0.53$, $p = 0.000$), T lymphocytes ($r = 0.5$, $p = 0.000$), cytotoxic T lymphocytes ($r = 0.48$, $p = 0.000$), Th17 lymphocytes ($r = 0.41$, $p = 0.003$) and NK cells ($r = 0.32$, $p = 0.023$). The concentration of aspartate aminotransferase (AST) increased with an increase in the number of activated cells with NF-κB translocation in the populations of T helpers ($r = 0.55$, $p = 0.000$), cytotoxic T lymphocytes ($r = 0.53$, $p = 0.000$), T lymphocytes ($r = 0.52$, $p = 0.000$), Th17 lymphocytes ($r = 0.28$, $p = 0.041$).

The concentration of gamma-glutamine transferase (GGT) increased with an increase in the number of activated cells with NF-κB translocation in the populations of T helper cells ($r = 0.54$, $p = 0.000$), T lymphocytes ($r = 0.52$, $p = 0.000$), cytotoxic T lymphocytes ($r = 0.5$, $p = 0.000$), Th17 lymphocytes ($r = 0.42$, $p = 0.0021$). Thus, an increase in the number of cells with NF-κB translocation in populations of T cell immunity may reflect the severity of the patient's condition, possibly by activating the inflammatory pathway involving NF-κB [13].

Considering the toxicological and inflammatory effects of copper described by Deng H. [2] and the ability of copper to influence the NF-κB signaling pathway, we investigated the relationship between urinary copper levels and NF-κB activity in lymphocyte populations. In children with WD, the copper concentration ranged from 9.7 to 2582 μg/day, Me = 616 (210-1173). We obtained a statistically significant direct relationship between the copper content in urine and the level of NF-κB translocation in B lymphocytes, $r = 0.34$, $p = 0.016$.

Conclusion

Thus, in most cases, children with WD are characterized by a decrease in the activity of the transcription factor NF-κB in populations of B cells, T helpers, T cytotoxic and Th17 lymphocytes. The activity of the factor correlates with biochemical markers of the severity of liver damage, as well as with the content of copper in the urine in children with WD. The study of the NF-κB signaling pathway seems to be promising for a better understanding of the pathogenetic mechanisms of the formation of liver inflammation and fibrosis in children with Wilson's disease.

References

1. Barteneva N.S., Vorobjev I.A. Imaging Flow Cytometry Methods and protocols. *Methods Mol. Biol., 2017, pp. 178-188.*

2. Deng H., Zhu S., Yang H., Cui H., Guo H., Deng J., Ren Z., Geng Y., Ouyang P., Xu Z., Deng Y., Zhu Y. Dysregulation of inflammatory pathways triggered by copper exposure. *Biol. Trace Elem. Res., 2023, Vol. 201, no. 2, pp. 539-548.*

3. Fisenko A.P., Surkov N.N., Potapov A.S. Wilson's disease in children. Moscow: National Medical Research Center of Children's Health, 2019. P. 83. (In Russ.)

4. Kuptsova D.G., Petrichuk S.V., Murashkin N.N., Kurbatova O.V., Radygina T.V., Khotko A.A., Ivanov R.A. Activity of nuclear factor κB in lymphocyte populations of children with psoriasis. Bulletin of the Russian State Medical University, 2022, no. 2, pp. 28-35. (In Russ.)

5. Kurbatova O.V., Kuptsova D.G., Bezrukavnikova L.M., Radygina T.V., Movsisyan G.B., Anushenko A.O., Komarova A.D., Potapov A.S., Petrichuk S.V., Fisenko A.P. Features of immune status in children with Wilson–

Konovalov disease at different stages of liver fibrosis. *Russian Journal of Immunology, 2022, Vol. 25, no. 4, pp. 453-460.* (In Russ.)

6. Lucena-Valera A., Ruz-Zafra P., Ampuero J. Wilson's disease: overview. Enfermedad de Wilson. *Med. Clin. (Barc.), 2023, Vol. 160, no. 6, pp. 261-267.*

7. Luedde T., Schwabe R.F. NF-κB in the liver-linking injury, fibrosis and hepatocellular carcinoma. *Nat. Rev. Gastroenterol. Hepatol., 2011, Vol. 8, no. 2, pp. 108-118.*

8. Mehto S., Kundu S., Chauhan S., Chauhan S. RIPosomes are targets of IRGM-SQSTM1-dependent autophagy. *Autophagy, 2023, Vol. 19, no. 3, pp. 1045-1047.*

9. Oakley F., Mann J., Nailard S., Smart D., Mungalsingh N., Constandinou C., Ali S., Wilson S.J., Millward-Sadler H., Iredale J.P., Mann D.A. Nuclear factor-kappaB1 (p50) limits the inflammatory and fibrogenic responses to chronic injury. *Am. J. Pathol., 2005, Vol. 166, no. 3, pp. 695-708.*

10. Petrichuk S.V., Radygina T.V., Kuptsova D.G., Kurbatova O.V., Semikina E.L., Murashkin N.N., Potapov A.S., Fisenko A.P. Evaluation of anti-TNF treatment efficiency in children with immune-dependent diseases by means of testing the NF-κB activity in lymphocyte populations. *Russian Journal of Immunology, 2022, Vol. 25, no. 4, pp. 491-498.* (In Russ.)

11. Petrichuk S., Radigina T., Gerasimova D., Illarionov A., Anushenko A., Erlikh-Fox T., Potapov A., P070 The level of nuclear factor kappa B (NF-kB) translocation during infliximab therapy in children with IBD. *J. Crohns Colitis, 2019, Vol.13, Iss. suppl. 1, pp. S120-S121.*

12. Przybyłkowski A., Szeligowska J., Januszewicz M., Raszeja-Wyszomirska J., Szczepankiewicz B., Nehring P., Górnicka B., Litwin T., Członkowska A. Evaluation of liver fibrosis in patients with Wilson's disease. *Eur. J. Gastroenterol. Hepatol., 2021, Vol. 33, no. 4, pp. 535-540.*

13. Zhang T., Hu J., Wang X., Zhao X., Li Z., Niu J., Steer C.J Zheng G., Song G. MicroRNA-378 promotes hepatic inflammation and fibrosis via modulation of the NF-κB-TNFα pathway. *J Hepatol., 2019, Vol. 70, no. 1, pp. 87-96.*

Авторы:

Курбатова О.В. *— к.м.н., старший научный сотрудник лаборатории экспериментальной иммунологии и вирусологии ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ, Москва, Россия*

Петричук С.В. *— д.б.н., профессор, главный научный сотрудник лаборатории экспериментальной иммунологии и вирусологии ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ, Москва, Россия*

Купцова Д.Г. *— младший научный сотрудник, врач клинической лабораторной диагностики лаборатории экспериментальной иммунологии и вирусологии ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ, Москва, Россия*

Мовсисян Г.Б. *— к.м.н., старший научный сотрудник лаборатории редких наследственных болезней, врач-гастроэнтеролог гастроэнтерологического отделения с гепатологической группой ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ, Москва, Россия*

Authors:

Kurbatova O.V., *PhD (Medicine), Senior Research Associate, Laboratory of Experimental Immunology and Virology, National Medical Research Center of Children's Health, Moscow, Russian Federation*

Petrichuk S.V., *PhD, MD (Biology), Professor, Chief Research Associate, Laboratory of Experimental Immunology and Virology, National Medical Research Center of Children's Health, Moscow, Russian Federation*

Kuptsova D.G., *Junior Research Associate, Clinical Laboratory Physician, Laboratory of Experimental Immunology and Virology, National Medical Research Center of Children's Health, Moscow, Russian Federation*

Movsisyan G.B., *PhD (Medicine), Senior Research Associate, Laboratory of Rare Hereditary Diseases, Gastroenterologist of the Gastroenterology Department with the Hepatological Group, National Medical Research Center of Children's Health, Moscow, Russian Federation*

Радыгина Т.В. *— к.м.н., старший научный сотрудник лаборатории экспериментальной иммунологии и вирусологии ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ, Москва, Россия*

Radygina T.V., *PhD (Medicine), Senior Research Associate, Laboratory of Experimental Immunology and Virology, National Medical Research Center of Children's Health, Moscow, Russian Federation*

Комарова А.Д. *— врач-педиатр гастроэнтерологического отделения с гепатологической группой ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ, Москва, Россия*

Komarova A.D., *Paediatrician, Gastroenterology Department with the Hepatological Group, National Medical Research Center of Children's Health, Moscow, Russian Federation*

Анушенко А.О. *— врач-гастроэнтеролог гастроэнтерологического отделения с гепатологической группой ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ, Москва, Россия*

Anushenko A.O., *Gastroenterologist of the Gastroenterology Department with the Hepatological Group, National Medical Research Center of Children's Health, Moscow, Russian Federation*

Фрейдлин Е.В. *— лаборант-исследователь лаборатории экспериментальной иммунологии и вирусологии ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ, Москва, Россия*

Freidlin E.V., *Laboratory Assistant, Laboratory of Experimental Immunology and Virology, National Medical Research Center of Children's Health, Moscow, Russian Federation*

Семикина Е.Л. *— д.м.н., руководитель лабораторного отдела ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ; профессор кафедры педиатрии и детской ревматологии ФГАОУ ВО «Первый Московский государственный медицинский университет имени И.М. Сеченова» Министерства здравоохранения РФ (Сеченовский университет), Москва, Россия*

Semikina E.L., *PhD, MD (Medicine), Head, Laboratory Department, National Medical Research Center of Children's Health; Professor, Department of Pediatrics and Pediatric Rheumatology, I. Sechenov First Moscow State*

Потапов А.С. *— д.м.н., профессор, главный научный сотрудник лаборатории научных основ детской гастроэнтерологии и гепатологии, заведующий гастроэнтерологическим отделением с гепатологической группой ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ; профессор кафедры педиатрии и детской ревматологии ФГАОУ ВО «Первый Московский государственный медицинский университет имени И.М. Сеченова» Министерства здравоохранения РФ (Сеченовский университет), Москва, Россия*

Potapov A.S., *PhD, MD (Medicine), Professor, Chief Research Associate, Laboratory of Scientific Foundations of Pediatric Gastroenterology and Hepatology, Head of Gastroenterology Department with Hepatology Group, National Medical Research Center of Children's Health; Professor, Department of Pediatrics and Pediatric Rheumatology, I. Sechenov First Moscow State Medical University (Sechenov University), Moscow, Russian Federation*

Фисенко А.П. *— д.м.н, профессор, директор ФГАУ «Национальный медицинский исследовательский центр здоровья детей» Министерства здравоохранения РФ, Москва, Россия*

Fisenko A.P., *PhD, MD (Medicine), Professor, Director, National Medical Research Center of Children's Health, Moscow, Russian Federation*

Поступила 14.04.2023
Отправлена на доработку 19.04.2023
Принята к печати 20.04.2023

Received 14.04.2023
Revision received 19.04.2023
Accepted 20.04.2023