Медицинская иммунология
2019, Т. 21, № 1, стр. 21-38

Обзоры
Reviews

Medical Immunology (Russia)/
Meditsinskaya Immunologiya
2019, Vol. 21, № 1, pp. 21-38

ДИФФЕРЕНЦИРОВКА NK-КЛЕТОК. ВЗГЛЯД ЧЕРЕЗ ПРИЗМУ ТРАНСКРИПЦИОННЫХ ФАКТОРОВ И ВНУТРИКЛЕТОЧНЫХ МЕССЕНДЖЕРОВ

Михайлова В.А.[1, 2], Баженов Д.О.[1], Белякова К.Л.[1], Сельков С.А.[1], Соколов Д.И.[1, 2, 3]

[1] ФГБНУ «Научно-исследовательский институт акушерства, гинекологии и репродуктологии имени Д.О. Отта», Санкт-Петербург, Россия
[2] ГБОУ ВПО «Первый Санкт-Петербургский государственный медицинский университет имени академика И.П. Павлова» Министерства здравоохранения РФ, Санкт-Петербург, Россия
[3] ФГБНУ «Институт экспериментальной медицины», Санкт-Петербург, Россия

Резюме. Все лимфоидные клетки относят к врожденному или адаптивному звену иммунитета согласно механизмам реализации иммунных реакций. Реализация функциональной активности NK-клеток не связана с процессами предварительной активации в результате контакта с антигеном, реаранжировкой генов антигенраспознающих рецепторов и клональной пролиферацией. В связи с этим NK-клетки традиционно относят к клеткам врожденного иммунитета. Ранее полагали, что единственной популяцией лимфоидных клеток врожденного иммунитета являются NK-клетки, однако в последние годы в литературе появляется все больше свидетельств существования различных популяций этих клеток, что послужило основанием для выделения общего кластера под названием «лимфоидные клетки врожденного иммунитета» (Innate Lymphoid Cells – ILC). По классификации ILC NK-клетки относят к первой группе врожденных лимфоидных клеток согласно их общим функциональным характеристикам, а также участию транскрипционного фактора T-bet в их дифференцировке. Сложность, многоэтапность и частично нелинейный характер дифференцировки NK-клеток связаны с влиянием клеточного микроокружения, последовательной экспрессией транскрипционных факторов и активацией различных внутриклеточных путей передачи сигнала в NK-клетках. В обзоре рассматривается место NK-клеток в классификации ILC, основные транскрипционные факторы, участвующие в процессе дифференцировки NK-клеток. Авторами предпринимается попытка обобщить основные пути внутриклеточной передачи сигнала в NK-клетках в зависимости от активации цитокинами, присутствующими в клеточном микроокружении и оказывающими влияние на NK-клетки. Особым объектом дифференцировки NK-клеток являются NK-клетки децидуальной оболочки при беременности. Кроме NK-клеток, в составе децидуальной оболочки присутствуют стромальные клетки, клетки трофобласта, макрофаги. В обзоре рассмотрен частный случай влияния клеток микроокружения на экспрессию транскрипционных факторов и активацию внутриклеточных мессенджеров NK-клеток на примере клеток трофобласта. Открытое в настоящее время многообра-

Адрес для переписки:
Михайлова Валентина Анатольевна
ФГБНУ «Научно-исследовательский институт акушерства, гинекологии и репродуктологии имени Д.О. Отта»
199034, Россия, Санкт-Петербург,
Менделеевская линия, 3.
Тел.: 8 (812) 328-98-50.
E-mail: mva_spb@mail.ru

Address for correspondence:
Mikhailova Valentina A.
D. Ott Research Institute of Obstetrics, Gynecology and Reproductology
199034, Russian Federation, St. Petersburg,
Mendeleev Line, 3.
Phone: 7 (812) 328-98-50.
E-mail: mva_spb@mail.ru

Образец цитирования:
В.А. Михайлова, Д.О. Баженов, К.Л. Белякова, С.А. Сельков, Д.И. Соколов «Дифференцировка NK-клеток. Взгляд через призму транскрипционных факторов и внутриклеточных мессенджеров» // Медицинская иммунология, 2019. Т. 21, № 1. С. 21-38. doi: 10.15789/1563-0625-2019-1-21-38

For citation:
V.A. Mikhailova, D.O. Bazhenov, K.L. Belyakova, S.A. Selkov, D.I. Sokolov "Differentiation of NK cells. A look through the prism of transcription factors and intracellular messengers", Medical Immunology (Russia)/Meditsinskaya Immunologiya, 2019, Vol. 21, no. 1, pp. 21-38. doi: 10.15789/1563-0625-2019-1-21-38

DOI: 10.15789/1563-0625-2019-1-21-38

зие NK-клеток, реализующееся в зависимости от микроокружения в процессе их дифференцировки, требует дальнейшего изучения.

Работа поддержана грантом НШ-2873.2018.7. Участие Баженова Д.О. поддержано стипендией Президента РФ СП-2836.2018.4.

Ключевые слова: NK-клетки, лимфоидные клетки врожденного иммунитета, дифференцировка, транскрипционные факторы, внутриклеточные мессенджеры, децидуальные NK-клетки, трофобласт

DIFFERENTIATION OF NK CELLS. A LOOK THROUGH THE PRISM OF TRANSCRIPTION FACTORS AND INTRACELLULAR MESSENGERS

Mikhailova V.A.[a, b], Bazhenov D.O.[a], Belyakova K.L.[a], Selkov S.A.[a], Sokolov D.I.[a, b, c]

[a] *D. Ott Research Institute of Obstetrics, Gynecology and Reproductology, St. Petersburg, Russian Federation*
[b] *First St. Petersburg State I. Pavlov Medical University, St. Petersburg, Russian Federation*
[c] *Institute of Experimental Medicine, St. Petersburg, Russian Federation*

Abstract. All lymphoid cells are referred to as an innate or adaptive immunity unit in terms of the mechanisms of performing immune reactions. The functional activity of natural killer (NK) cells is not associated with pre-activation processes resulting from contact with antigen, rearrangement of antigen-recognition receptor genes, and clonal proliferation. In this regard, NK cells are traditionally referred to as cells of innate immunity. Previously, it was believed that NK cells represent the only population of innate immunity lymphoid cells, but, more recently, there has been increasing evidence in the literature concerning existence of different populations of these cells, thus serving a basis for isolating a common cluster called Innate Lymphoid Cells (ILC). According to the ILC classification, NK cells are classified as the first group of innate lymphoid cells according to their overall functional characteristics, as well as contribution of the T-bet transcription factor to their differentiation. Complexity, multistage and partially nonlinear character of NK cell differentiation are associated with influence of the cellular microenvironment, consistent expression of transcription factors and activation of various intracellular signaling pathways in NK cells. The review considers positioning of NK cells in the ILC classification, the main transcription factors involved in NK cell differentiation. The authors are seeking for generalization of the major routes of intracellular signal transmission in NK cells depending on their activation by cytokines located in the cellular microenvironment and affecting NK cells. The decidual NK cells during pregnancy represent a special object of NK cell differentiation. Stromal cells, trophoblast cells and macrophages are present in the decidua, in addition to NK cells. The review concerns a special case of microenvironmental effects upon expression of transcription factors and activation of NK intracellular messengers, while considering trophoblast cells an example of such influences. The recently discovered variety of NK cells, induced by the microenvironment in the course of their differentiation, requires further study. The work is supported by the grant NSh-2873.2018.7. The participation of D.O. Bazhenov was supported by a Presidential scholarship of the Russian Federation SP-2836.2018.4.

Keywords: NK cells, innate immunity lymphocytes, differentiation, transcription factors, intracellular messengers, decidual NK cells, trophoblast

The work was supported by the Council on Grants of the President of the Russian Federation (Grant No. NSh-2873.2018.7). The participation of D.O. Bazhenov was supported by a scholarship of the President of the Russian Federation (SP-2836.2018.4).

Lymphoid cells of innate immunity

All lymphoid cells are referred to as an innate or adaptive immunity unit in terms of the mechanisms of performing immune reactions. Lymphoid cells that determine the development of reactions of adaptive immunity are T and B lymphocytes. In the process of their antigen-independent differentiation, the antigen-recognizing receptors of T and B lymphocytes (TcR and Ig, respectively) mature, including recombining the gene sections of the chains of these receptors. The biological meaning of recombination of the genes of T and B lymphocyte receptors is to increase the diversity of the antigen-recognition regions of the receptors. After recognition of the antigen by T and B lymphocytes, their clonal proliferation takes place, when a large number of lymphocytes are formed, capable of binding a specific antigen, which stimulated

their proliferation. Clonal proliferation increases the number of $CD4^+$T-helper lymphocytes, $CD8^+$ cytotoxic T and B lymphocytes [23]. During antigen-dependent differentiation of T and B lymphocytes, part of the proliferating clone cells differentiates as memory cells [23]. Among lymphoid cells, controlling the development of effector mechanisms of innate immunity, NKT cells and NK cells are determined [23]. Implementation of the functional activity of NK cells is neither associated with the processes of pre-activation due to the contact with antigen, nor with the rearrangement of gene antigen-recognizing receptors, nor with the clonal proliferation; and therefore the NK cells are traditionally attributed to the cells of innate immunity [23].

Previously, it was believed that NK cells are the only population of innate immunity lymphoid cells. However, in recent years, there has been increasing evidence in the literature on the existence of different populations of lymphoid cells that belong to the group of innate immunity lymphoid cells (Innate Lymphoid Cells – ILC). Most of the data on the characteristics of ILC, presented in the literature, was obtained by studying them in model animals, mainly mice. Among the ILC, conventional NK cells (cNK) and ILC1, ILC2 and ILC3 populations are distinguished. In 2013, Spits et al. proposed the nomenclature of lymphoid cells of innate immunity, based on the cytokines secreted by these cells and the main transcription factors involved in their differentiation [68]. They identified three groups of ILC. The cells of the first group (group 1 ILC) include ILC1 and NK cells secreting IFNγ [68]. For group 1 ILC, namely, ILC1 and NK cells, expression of the T-bet transcription factor is characteristic. At the same time, in addition to T-bet, NK cells also express the Eomes transcription factor [69]. The cells of the second group of ILC (group 2 ILC) are ILC2, which are characterized by the secretion of IL-5 and IL-13. Differentiation of ILC2 occurs with the participation of the RORα and GATA3 transcription factors [68]. The third ILC group (group 3 ILC) is ILC3, for the differentiation of which RORγt is required, and which secrete IL-17 and IL-22. Within group 3 ILC, ILC3 population is distinguished, which secrete only IL-22, and when stimulated with IL-12 and IL-18, may differentiate as ILC1, as well as lymphoid tissue-inducing cells (LTi), necessary for lymph node formation during the embryogenesis stage [68]. In humans, individual ILC groups (shown for $RORc^+$ILC, which can be classified as ILC3) may be in an inactive state in secondary lymphoid organs, but when activated may be involved in the implementation of inflammatory responses due to IL-17 and IL-22 cytokine production. [30]. For the differentiation of all ILC groups, expression of the cytokine common γ-chain is required, which is a part of IL-2R, IL-7R, IL-9R, IL-15R, IL-21R [68] (Table 1). Given the diversity of ILC and certain similarities to existing groups of T-helper (Th) lymphocytes, existence is possible of ILC evolutionary continuity between innate immunity and T lymphocytes of the adaptive immunity [70, 73].

Analysis of transcribed genes in ILC1, ILC2, ILC3 and NK cells did not show unique genes that regulate the development of a particular population [56], indicating flexibility of individual stages of differentiation, and the possibility of mutual ILC transitions [56]. For example, ILC2 are characterized by secretion of IL-5 and IL-13 after stimulation with IL-25 or IL-33 [18], while ILC3 are characterized by the expression of IL-22 and IL-17 [18]. Between the populations of ILC2 and ILC3, there is the possibility of mutual transition: under the influence of IL-12, ILC2

TABLE 1. CHARACTERISTICS OF ILC GROUPS

Characteristic	cNK	ILC1	ILC2	ILC3
Phenotype (in humans)	$CD3^-CD56^+$ [69] $CD4^-$ [68] $NKp46^+$ [69] $CD49b^+$ [69] $CD94^+$ [69] $NKG2D^+$ [69] $NKp30^+$ [69] $NKp46^+$ [69] $IL\text{-}12R^+$ [66] $IL\text{-}15R^+$ [66] $IL\text{-}18R^+$ [66]	$CD3^-$ [28] $CD4^-$ [68] $CD27^+$ [69] $CD56^-$ [69] $CD94^-$ [69] $CD127^+$ [69] $NKp46^-$ [68]	$CD3^-$ [28] $CD4^-$ [68] $CD127^+$ [68] $NKp46^-$ [68] $CRTH2^+$ [66] $IL\text{-}18R^+$ [66] $CD161^+$ [66] $IL\text{-}25R^+$ [66] $IL\text{-}33R^+$ [66] $IL\text{-}17R^+$ [28]	$CD3^-$ [28] $CD4^-$ [68] $CD56^-$ [69] $CD127^+$ [68] $NKp44^+$ [66] $IL\text{-}18R^+$ [66] $IL\text{-}1R^+$ [66] $IL\text{-}23R^+$ [66]
Secreted cytokines	IFNγ [68] TNFα [66]	IFNγ [68] TNFα [66]	IL-5, IL-13 [68] IFNγ [66] IL-4 [66] IL-3, IL-8 [28] IL-9, IL-21 [28]	IL-17, IL-22 [66, 68] IL-2, TNFα [66]
Transcription factors	T-bet [69] Eomes [69]	T-bet [69]	RORα [68] GATA3 [68]	RORγt [68]

and ILC3 can be transdifferentiated into ILC1. Under the influence of IL-4 and IL-23, ILC1 in turn can differentiate into ILC2, and IL-23 will promote differentiation of ILC1 into ILC3 [69]. It was shown that LTi can differentiate into ILC2 and secrete IL-5 and IL-13 after exposure to IL-2 in combination with TLR2 ligands [13]. Such flexibility is associated with inhibition of GATA-3 and RORγt transcription factors and the activation of T-bet [69]. It was shown that RORγt$^+$ ILC can differentiate into ILC1 under the influence of IL-2 and IL-12 [4]. Depending on the tissue (thymus, liver, salivary glands), the phenotype of ILC1 and their functional characteristics may differ. For example, the liver ILC1 are characterized by the performance of cytotoxicity through the TRAIL-mediated pathway, and not through granzymes and perforin [69]. Probably, the conditions of the microenvironment can have a significant effect on the ILC, erasing to a certain extent the differences between the currently determined ILC1 and cNK [60, 69].

Not only is the differentiation of ILC under the influence of the cytokine microenvironment, but vice versa, ILC affect the surrounding cells and tissues. So, ILC of the spleen, located near the T and B lymphocytes, in the adult body express the OX40L and CD30L costimulatory molecules, characteristic for T lymphocytes. Due to these costimulatory molecules, the spleen ILC can contribute to the survival of Th2 lymphocytes and memory T cells [70]. ILC promote differentiation of plasma cells in Peyer's glands and switching of synthesized antibodies towards IgA [70]. In the reviews of Tait Wojno et al. (2012) and Spits et al. (2012), the authors describe that ILC2, under the influence of IL-25 and IL-33 secreted by intestinal epithelial cells in helminthiasis, increase the secretion of IL-5, which stimulates the proliferation of eosinophils. Also, ILC2 support activation of Th2 lymphocytes and stimulate, thus, the adaptive immune response [70, 72]. Thus, the formation of different ILC groups is performed under the control of microenvironment cells. At the same time, ILC can influence both other cells of innate immunity, and regulate the reactions of adaptive immunity.

Place of NK cells in the group of lymphoid cells of innate immunity

cNK cells are considered in the framework of differentiation of the first group of ILC, which also include ILC1. At the moment, it has not been possible to identify intracellular and surface markers that would clearly allow the population of cNK cells and ILC1 to be separated (Table 1) [69]. A significantly overlapping spectrum of transcribed genes in NK cells and ILC1 suggests both the late discrepancy between these populations in the differentiation process and the subsequent differentiation of immature NK cells and later mature NK cells, from ILC1 [56].

Despite the existence of a generalizing classification of H. Spits et al., more and more refinements are observed in the literature concerning different ILC groups and the position of NK cells in this classification. In addition to the cNK cells present predominantly in the peripheral blood, nonconventional NK cells are distinguished within the NK cell population, which in turn include the NK cells of the thymus, liver, lungs, skin, uterus, kidneys, pancreas and salivary glands [20]. The existing diversity of populations of tissue-resident NK cells causes difficulties with the unambiguous determination of their place in the ILC classification. In experiments using mice, it has been shown that, in the absence of infections, ILC1 remains predominantly tissue-resident, while cNK cells recirculate [69]. In some works, cNK is referred to ILC with predominantly killer activity [18]. At the same time, in other studies, ILC1 is differentiated from cNK cells by the reduced or no cytotoxic activity [69], the absence of expression of the *Gzma* and *Prf1* genes encoding, respectively, granzyme A and perforin [56], absence of granzyme B and perforin [4], as well as the absence of expression of receptors for IL-15 and KIR3DL1 [4]. At the same time, the separation of ILC1 and cNK is to a certain extent conditional, since in part the expression profiles of surface receptors of these cell populations [18], as well as their transcription factors [68] overlap. Thus, analysis of transcriptomic programs of ILC and NK cells revealed that NK cells are different from ILC1 by the presence of the *Eomes* gene expression in the liver and in the spleen and in the lamina propria of the mouse small intestine [56]. In general, the mouse model shows that the *Tnfsf10*, *Tnf* and *Cxcr6* genes are transcribed in the spleen and liver ILC1, while the *Eomes* genes, *Itgam* genes (encoding CD11b), the *Klra* gene family (encoding the Ly49 family) are transcribed in the NK cells [56].

Thus, according to the ILC classification, NK cells belong to the group 1 innate lymphoid cells according to their general functional characteristics, as well as the participation of the T-bet transcription factor in their differentiation. The complexity and multistage process of differentiation of NK cells involves the participation of many transcription factors, which will be discussed in the next section.

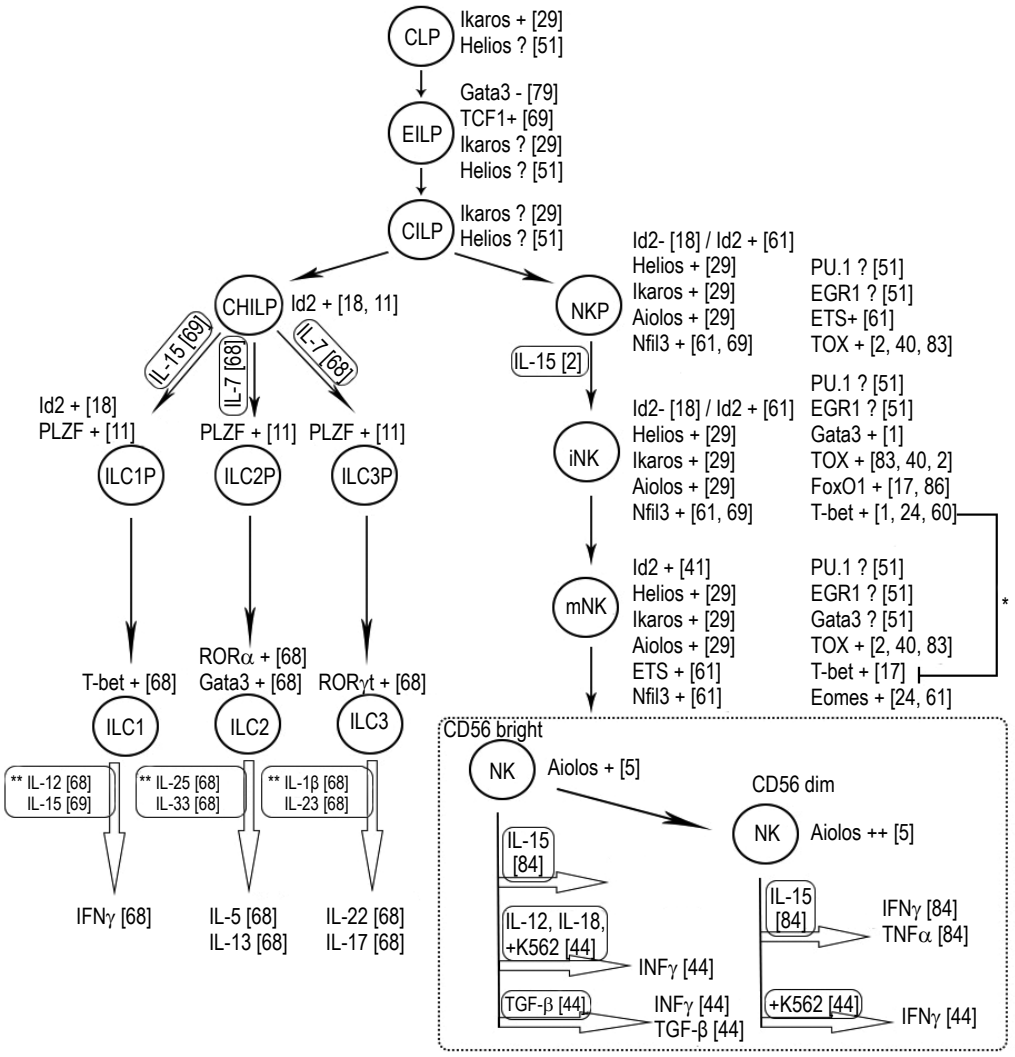
Participation of transcription factors in the process of differentiation of NK cells within the ILC branch

The mouse is the model object in most studies concerning the research of transcription factors involved in the control of differentiation of various ILC groups, in particular NK cells. Despite certain differences in the ILC phenotype in humans and mice, we may make some analogy, since the ILC localization, their functions and the transcription factors participating in their formation, are similar in the human and mouse ILC [72]. Differentiation of different stages of NK cells involves a spectrum of different transcription

factors: Id2, Nfil3, TOX, ETS1, Eomes, GATA-3, FoxO1, Ikaros, T-bet, Aiolos [29, 53, 60, 83]. However, these transcription factors are not unique to the regulation of NK cells, they also participate in the differentiation of other ILCs. At present, the role of these factors in the differentiation of NK cells is not yet defined unambiguously. This section provides information on some of the transcription factors, which impact the main stages of maturation of NK cells (Figure 1).

Id2 transcription factor in differentiation of NK cells

Forming the bulk of NK cells occurs in the bone marrow, but NK cells may also differentiate in the lymph nodes of $CD34^+$ hematopoietic stem cells [45, 59]. It is believed that the differentiation of NK cells begins with the stage of the common lymphoid progenitor (CLP) [18]. CLP differentiates into an early progenitor of ILC (EILC), the formation of which is associated with the expression of the transcription factor TCF1 [69]. Sequential differentiation into the common progenitor of ILC (CILP) is associated with the expression of the Id2 transcription factor [18]. Id2 can also claim to be the main transcription factor regulating ILC differentiation, since mice with a defect in this transcription factor were characterized by the absence of all ILC populations, while the differentiation of T and B lymphocytes was not impaired [18]. If Id2 is defective, mice do not develop lymphoid organs and Peyer's glands, and the number of NK cells is greatly reduced [89]. The Id2 transcription factor is one of



* – FoxO1 suppresses the expression of the T-bet transcription factor [17].

** – Due to the diversity of ILC, depending on the tissue microenvironment, the cytokines regulating the differentiation of individual ILC populations are not clearly defined.

? – Differences in the tactics used to investigate the degree of differentiation of NK cells complicate the unambiguous attribution of these factors to a certain stage in the presented scheme.

Figure 1. The main stages of differentiation of NK cells within the ILC classification and associated transcription factors

Note. CLP is a common lymphoid progenitor, EILC is an early progenitor of ILC, CILP is a common progenitor of ILC, CHILP is a common lymphoid progenitor of innate lymphocytes similar to helper cells, ILC1P is an ILC1 progenitor, ILC2P is an ILC2 progenitor, ILC3P is an ILC3 progenitor, NKP is a progenitor of NK cells, iNK are immature NK cells, mNK are mature NK cells.

the proteins that bind to E-proteins, thus blocking the possibility of binding of E-proteins and DNA [70]. E-proteins include E12 and E47, which regulate the development of B lymphocytes and participate in the differentiation of T lymphocytes [70]. E-proteins also include E2-2, which determines the development of plasmacytoid dendritic cells, and HEB involved in the development of T lymphocytes [70]. Thus, Id2 controls the switching of the CLP differentiation program towards ILC [70]. Later, $Id2^+$ CILP was isolated as a separate stage of differentiation, a common lymphoid progenitor of innate lymphocytes like helper cells (CHILP), which in turn gives rise to differentiation of various ILC populations, but not cNK [69]. $Id2^+$ CHILP are not a homogeneous population by the profile of transcription factors, since, for example, only 50% of them express the PLZF transcription factor, encoded by the *Zbtb16* gene [11]. Previously, this transcription factor was considered as determining the differentiation of NKT cells [11]. $PLZF^{high}$ + ILC progenitors give rise to ILC1, ILC2, ILC3, but not NK cells [11]. The CHILP stage in ILC differentiation has been validated for mice; for humans, the CILP-ILC transition linearity and presence of the CHILP stage is under discussion [59]. Data on Id2 expression after the CILP stage are inconsistent. According to one data, after the CILP stage, Id2 is not expressed, since the progenitors of NK cells are predominantly Id2- [18]. According to other data, NKP weakly express Id2 [61]. In the late stages of differentiation, the mature NK cells (mNK) express Id2 [18]. Probably, Id2 is expressed in immature NK cells (iNK), since in experiments using mice it was shown that Id2 is involved in the stage of differentiation of immature NK cells (iNK) into mature NK cells (mNK) [40]. In mice, it was shown that as the NKP differentiates into mNK, Id2 expression increases [61]. In the model of differentiation of human NK cells from $CD34^+$ mononuclear cord blood cells *in vitro*, it was shown that in addition to Id2, in the early stages of NK cell line differentiation, transcription factors EGR-1 and PU.1 are expressed, then their expression decreases [51]. The cell phenotype after different cultivation times in this research was not evaluated, which makes it difficult to accurately determine the stage of differentiation where these transcription factors were expressed.

GATA-3 transcription factor in differentiation of NK cells

The differentiation of NK cells is negatively affected by the activation of the transcription factor GATA-3 at the CLP stage, which leads to disruption in the formation of NK cells and the predominance of T lymphocyte differentiation. Inhibition of GATA-3, regulating Notch signaling pathway in CLP, promotes transcription of genes involved in controlling formation of NK cells [79]. At the same time, the mouse model shows that the deficiency of GATA-3 in the later stages of differentiation of NK cells, namely iNK, leads to a decrease in the expression of *Id2*, *T-bet* and *Nfil3 (E4bp4)* genes, which in turn leads to disrupting differentiation of NK cells at the early stages [1]. For example, NK cells of mice deficient of GATA-3 showed reduced ability to secrete IFNγ, however degranulation and associated expression of LAMP-1 by NK cells was comparable with that of intact control mice [1]. Also, migration from the bone marrow has been impaired in GATA-3-NK cells, which is probably due to a defect in the expression of the CD11b adhesion molecule and an increased expression of CXCR4 binding the chemokine CXCL12 present in the bone marrow [1]. At the same time, iNK with GATA-3 defect has a high proliferative ability, when infected with mouse cytomegalovirus (MCMV), their number increases. Given that the cytotoxic activity of the cells was maintained at a level comparable to the cytotoxicity of intact NK cells, it's possible that iNK may participate in the antiviral defense [1]. The transcription factor GATA-3 not only participates in the regulation of the early stages of differentiation of NK cells. For humans, the expression of *GATA-3* is accompanied by reduced expression of *NKG2A* gene which indicates a GATA-3 involvement in the regulation of the functional properties of NK cells [51].

Transcription factor ETS1 in differentiation of NK cells

In mice, it was shown that the differentiation of the NK cell line is associated with the expression of the transcription factor ETS1 predominantly in the iNK [52]. According to Seillet et al., *ETS1* expression is observed in the stages NKP, iNK and mNK with peak at the stage of iNK [61]. ETS1 binds directly to the *Tbx21* gene, which encodes the transcription factor T-bet, and *Cd122,* which encodes the IL-2 receptor, and regulates the expression of Id2 [52]. It should be noted that, despite the involvement of the ETS1 transcription factor in the early stages of NK cell differentiation, the knockout of its gene results in a violation of the expression by NK cell of such functional receptors as NKp46, Ly49D, Ly49H in mice, a proliferation disorder in response to IL-15 and violation of the cytotoxic activity of NK cells associated with the release of granzymes and perforin [52].

Transcription factor Nfil3 (E4bp4) in differentiation of NK cells

In a study in mice, it was also shown that the mRNA of the transcription factor Nfil3 (E4bp4) was detected in the stages of differentiation of NKP, iNK and mNK [61]. An increase in the expression of the *Nfil3* gene was observed in mNK [41]. In mice having a mutant *Nfil3* gene, decreased expression of *Eomes* was observed in mNK, indicating the involvement of the transcription factor Nfil3 in regulating differentiation of mature NK cells [61]. Nfil3 can bind to the regulatory regions of DNA controlling transcription of Id2 and Eomes [41]. Expression of *Nfil3* and *Id2*

transcription factors in NKP cells leads to a synthesis and exposure on the cell surface of the IL-15 receptor (IL-15Rβ/γ) encoded by *Il2rb* and *Il2rg* genes [53]. In this way, Nfil3 promotes the differentiation of mature NK cells by inducing Eomes expression [41, 61]. However, the expression of Nfil3 is not homogeneous for different populations of tissue-resident NK cells. It has been found that NK cells and ILC1 of the lamina propria of the mouse small intestine stand out by most highly expressed *Nfil3* gene, while low expression of this factor is characteristic of NK cells and ILC1 of liver and spleen [56].

Transcription factors of the Ikaros family in differentiation of NK cells

During differentiation of NK cells from CLP, expression of *Ikzf1* and *Ikzf2* genes was also observed, which encode, respectively, transcription factors of the Ikaros family (Ikaros and Helios) [29, 51]. Moreover, *Ikzf1* expression persists throughout the whole path from CLP to mNK [29]. Expression of *Ikzf2* is low at the initial stages of differentiation, when the NK cell line is not yet expressed (CLP), and increases in NKP, iNK and mNK [29]. In mice, it is shown that while the NKP line is isolated, the expression of the *Ikzf3* gene encoding the transcription factor Aiolos, which also belongs to the Ikaros factor family, also occurs. And its expression is preserved until the stage mNK [29]. Aiolos regulates differentiation of NK cells regardless of the factor T-bet [29]. In mice having a mutant *Ikzf3* gene, presence of large amounts of immature NK cells in bone marrow was observed [29]. The transcription factor Aiolos is also involved in the regulation of the functional activity of mature NK cells. In mice with a gene knockout of this factor, NK cells possess increased cytotoxic activity against tumour cells, but did not cause the death of virus-infected cells [29]. In humans, Aiolos expression is shown for NK cells with phenotype $CD56^{dim}$ and $CD56^{bright}$ of peripheral blood, which can be regarded as mNK. In $CD56^{dim}$ NK cells, the Aiolos content was higher, which suggests the involvement of this factor in the terminal stages of differentiation of NK cells [5].

Transcription factor FoxO1 in differentiation of NK cells

In mice, it was shown that activation of transcription factor FoxO1 at the stage of iNK is necessary for differentiation of NK cells [17, 86]. This transcription factor regulates the autophagic process of cells, during which certain proteins and organelles degrade, and this ultimately helps to maintain cell homeostasis [86]. The process of autophagy promotes elimination of damaged mitochondria and thus prevents the induction of NK cells apoptosis caused by active oxygen species [86]. Mice knocked-out in FoxO1 demonstrate violations of the later stages of differentiation of NK cells and impaired homing of NK cells in the lymph nodes [17]. Mice knocked-out in FoxO1, also demonstrated increased cytotoxic activity against tumour cells, which indicates that FoxO1 has a moderating role in the anti-metastatic activity of NK cells [17]. Transcription factor FoxO1 is a negative regulator of differentiation of NK cells. When it is phosphorylated, the ability to bind to DNA is blocked. Cytokines such as IL-2, IL-12 and IL-15 induce phosphorylation of FoxO1, which was shown in NK cells, NK-92 line and murine spleen NK cells [17]. FoxO1 suppresses the expression of the transcription factor T-bet at the late stage of NK cell differentiation (mNK). With a FoxO1 defect, there is an increased production of T-bet, as well as an increase in the cytotoxic function of NK cells and the secretion of IFNγ [17].

Transcription factor TOX in differentiation of NK cells

In mice, it was shown that the transcription factor TOX is involved in differentiation from NKP to mNK [2, 40]. At the stage of iNK and mNK, *Tox* gene expression significantly exceeds the expression at the NKP stage [2]. In mice with a defect of this transcription factor, there is almost complete absence of LTi cells, as a result, the formation of lymph nodes is disrupted [2]. Mutants in the *Tox* gene of NK cells show a decreased production of Id2 [2]. For humans, there are two forms of the transcription factor – TOX1 and TOX2. The genes of both transcription factors were expressed in NK cells during differentiation from $CD34^+$ cells *in vitro*. Moreover, the *TOX1* and *TOX2* expression level increased with duration of cultivation and acquisition by cells of phenotype $CD34^-CD94^+$, which may indicate the involvement of these factors in regulating the formation of mature NK cells [83]. TOX2 can directly regulate the expression of the transcription factor T-bet [83]. By knockout of the *Tox* gene, the cytotoxic activity of NK cells is significantly reduced, which may be due to the observed reduced expression of perforin [83].

Transcription factors T-bet and Eomes in differentiation of NK cells

Transcription factors that are most frequently associated with terminal differentiation of NK cells are T-bet and Eomes [60, 75]. Mice show that T-bet regulates the formation iNK, stimulates NK cell egress from the bone marrow and induces the expression of IL-2Rβ [60]. Eomes contributes to the NK cells acquiring functional receptors Ly49 [60]. Also, using mice as the animal model demonstrated that violation of the T-bet expression results in disruption of maturation and differentiation of NK cells and loss of NK cells function associated with the antiviral response [55]. For NK cells of human peripheral blood, expression of T-bet and Eomes is also characteristic [65]. It should be pointed out, that one of the possible complications of allogeneic transplantation of hematopoietic stem cells is the reduction of antiviral

protection and the ability to prevent the "graft-versus-host" reaction by NK cells. This complication correlates with reduced expression of T-bet in NK cells, which in turn confirms the participation of T-bet in the implementation of the functional activity of NK cells in humans [65]. The mouse model shows that in the absence of the transcription factors T-bet and Eomes, the differentiation of NK cells is impaired. Eomes knocked-out mice revealed an increase of iNK cells with T-bet expression [24]. After stimulation of peripheral blood NK cells with IL-2, the expression of *Eomes* genes increases [25]. T-bet induces the expression of the transcription factor Zeb2, the activity of which promotes the release of NK cells from the bone marrow [80]. Zeb2 regulates terminal differentiation of NK cells in both mouse and human subjects. By increasing the expression of T-bet, Zeb2 expression is also increased, which is associated with terminal differentiation of NK cells [80].

Thus, during the differentiation of NK cells from CLP to mNK, a sequential regulation of the expression of transcription factors is observed, which in turn promotes the expression of receptors for cytokines and cytotoxic receptors, and also stimulates the synthesis of granzymes by NK cells. Partial regulation of transcription factors in the expression of receptors for cytokines determines the possibility of the effect of the cytokine microenvironment on the differentiation of NK cells. Cytokines of the microenvironment, by communicating with their corresponding receptors of NK cells, can activate various cascades of intracellular signaling. In the next section, the main cytokines involved in the regulation of NK cell differentiation and activation, as well as the signal cascades activated by them, will be examined.

Activation of intracellular signaling pathways in NK cells depending on the cytokines of the microenvironment

Microenvironment cells can influence both the differentiation of NK cells and their functional characteristics. For example, IL-15 and IL-2 stimulate the cytotoxic reactions of NK cells, while TGF-β suppresses them [27]. Depending on the cytokine, signaling in the NK cell can take place with the participation of different cascades of intracellular messengers.

Activation of intracellular signaling pathways in NK cells by cytokines which receptors contain a common γ chain: IL-15, IL-2, IL-7

IL-15 is one of cytokines that play an important part in the NK cells differentiation and acquisition of functional characteristics [16]. IL-15 refers to cytokines which receptors contain a common γ-chain (IL-2, IL-7), so that their intracellular signaling pathways may overlap. Depending on the target cells, IL-15 can trigger activation of the three intracellular signaling pathways from IL-15R: JAK1/JAK3/STAT3/STAT5; PI3K/Akt/mTOR; Ras/Raf/MAPK [7, 27, 46] (Figure 2). In the JAK-STAT pathway, several JAK-kinases are distinguished: JAK1, JAK2, JAK3 and TYK2, as well as STAT proteins 1, 2, 3, 4, 5A, 5B and 6 [27]. After the binding of IL-15 to its receptor IL-15Rβ/γ, the receptor-associated kinases JAK1 and JAK3 become active, which facilitates the phosphorylation of STAT proteins [53]. STAT1 is a key messenger starting IFNγ production by NK cells [27] and, together with STAT5, it is involved in the induction of activation and the cytotoxic activity of NK cells [25, 27]. Cytokine IL-15 induces phosphorylation of STAT3 [27]. It is shown that the intracellular messenger mTOR promotes the phosphorylation of STAT3 [42]. *STAT3* mutations cause increased NK cell proliferation, such mutations often are detected with NK cell lymphomas [36]. Similar mutations of *STAT3* promote enhanced phosphorylation of STAT5B, which consequently remains in the activated state for a long time [36]. In experiments in mice, it was shown that the absence of STAT3 in NK cells does not affect their differentiation, while enhancing the expression of granzyme B, perforin and the activating receptor DNAM-1. Thus, STAT3 inhibits the cytotoxic function of NK cells [26]. STAT5 participates in the signal transmission from IL-15 [27, 42]. For mouse NK cells, the secretion of VEGFA and EGF is shown, with a decrease in the synthesis of STAT5, an increase in the production of VEGFA and EGF is observed, which in turn stimulates the tumour growth [25]. Humans show high production of VEGFA by peripheral blood NK cells with the phenotype of CD16^{-}CD56^{bright}, together with a reduced amount of STAT5A and STAT5B [25]. The target gene of STAT5 is *Mcl1*, which maintains the survivability of the NK cell pool [53]. STAT5 is also involved in the regulation of the expression of anti-apoptotic genes *Bcl-xl, Bcl2* [14]. In mice having mutant *STAT5A* and *STAT5B* genes, there are no NK cells [14]. Activation of STAT6 reduces the cytotoxic activity of NK cells in cell culture [27]. A violation of the expression of the genes *Jak3, Stat5* or *Mcl1* leads to significant violations of NK cell functions [53].

When NK cells are activated via the JAK-STAT signaling pathway, the transcription of the *Cish* and *Socs1-7* genes occurs. The products of these genes (CIS and SOCS1-7, respectively) bind phosphorylated tyrosine kinase residues, and thus realize the mechanism of negative feedback, inhibiting the activation of NK cells [53]. The products of the genes *Cish* and *Socs1-7* belong to the SOCS family of proteins, the main mechanism of which is the initiation of proteasomal degradation of SOCS protein ligands [53]. CIS signaling protein binds JAK1, which phosphorylates STAT5, and triggers it proteasomal degradation, which leads to blocking of the signal from the receptor to IL-15 and containment thereby of the NK cells activation [15]. The mice with the *Cish* gene mutation observed increased proliferation of NK cells in re-

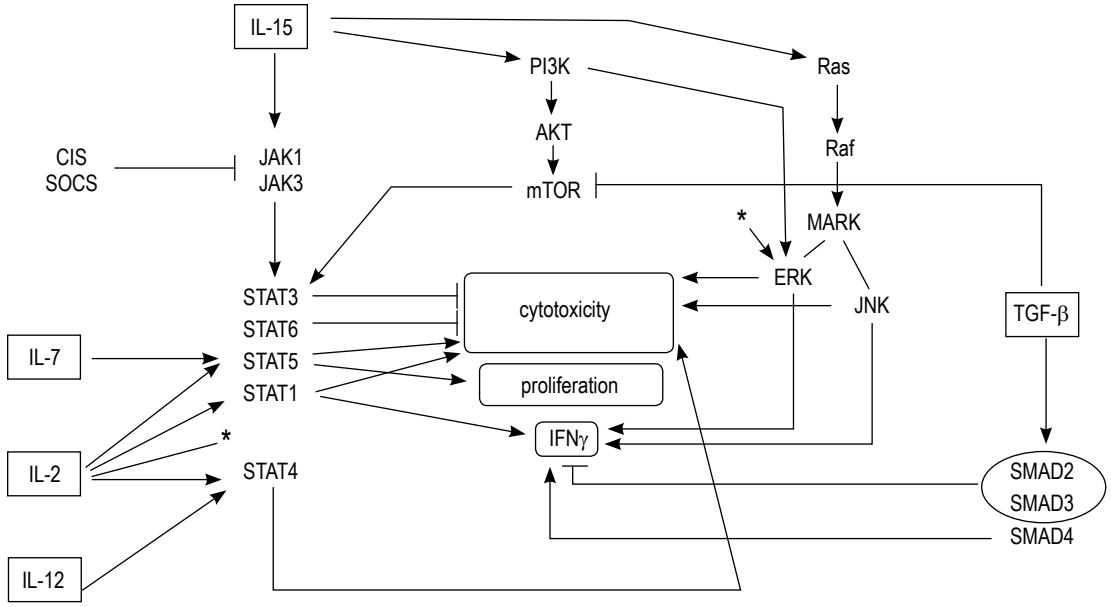


Figure 2. General scheme of activation of intracellular signaling pathways in NK cells depending on the microenvironment cytokines

Note. * IL-2 can cause activation of NK cells by phosphorylation of ERK.

sponse to low concentrations of IL-15 which did not cause proliferation of NK cells in the control group of mice [15]. In mice with a *Cish* gene mutation, no violations of AKT phosphorylation after IL-15 effects on NK cells were observed, indicating the specificity of blocking signal transmission from IL-15 by CIS protein precisely for the JAK-STAT pathway [15].

In addition to the JAK-STAT pathway for IL-15, activation of the MAPK (mitogen-activated protein kinase) signaling pathway, including activation of mitogen-associated protein kinases – ERK and JNK, is shown [49, 90]. Activation of these kinases by IL-2 cytokine, the receptor of which contains a common γ-chain with the IL-15 receptor, is accompanied by NK cells acquisition of the ability to secrete IFNγ and increase the cytotoxic activity [90]. As mentioned earlier, IL-15 also stimulates the signal through the signal pathway associated with the activation of mTOR [42], which participates in signal transmission from PI3K. So, in $CD56^{bright}$ NK cells, IL-15 causes activation of the PI3K/Akt/mTOR pathway, as well as the Ras/Raf/MAPK pathway of intracellular signaling. At the same time, during the IL-15 activation and induction of antitumor activity in the $CD56^{dim}$ NK cells, the PI3K/Akt/mTOR signaling pathway was not engaged [84]. It should be pointed that the PI3K/Akt/mTOR and Ras/Raf/MAPK paths can overlap. Thus, PI3K activation in NK cells leads to activation of ERK by Ras activation independent pathway that eventually leads to the induction of NK cell cytotoxicity [32]. For NK cells of the NK-92 line, regulation of signal transmission from the Syk kinase to the PI3K and ERK is also shown [32].

In addition to IL-15, activation of NK cells can be induced by IL-2 and IL-7. IL-2 has been shown to increase the proliferative activity of peripheral blood NK cells when administered intravenously to patients with advanced cancer [8] and when administered subcutaneously to patients with a chronic 'graft-versus-host' reaction. After intravenous administration of IL-2, NK cell secretion of IFNγ results in autocrine and paracrine activation of STAT1 NK cells and the formation of pSTAT1 [64]. In humans, at mutation of protein STAT1 gene, terminally differentiated NK cells as well as function of the cytotoxic NK cells are impaired [81]. In addition to STAT1, IL-2 induces activation of STAT4 in NK cells [85]. When the signal is transmitted from IL-2 and IL-7, activation of STAT5 occurs [27, 33]. In healthy donors with stimulation of IL-7 NK cells, pSTAT5 is formed, thereby increasing cytotoxicity of NK cells [33]. In chronic infections (HIV, hepatitis C virus) in NK cells, IL-7-induced phosphorylation of STAT5 is disrupted, which is associated with reduced cytotoxic function of NK cells [33].

The cytokine IL-2 can also cause activation of NK cells by phosphorylation of ERK1/2 [63]. In cell lines, it was shown that phosphorylation of ERK2 in NK cells leads to polarization of microtubules and cytolytic granules [10]. Cytokine IL-2 causes activation of surface receptors 2B4 and NKG2D, which in addition to activating ERK triggers the phosphorylation of kinase VAV-1 [77]. As a result, the induction of cytotoxicity is resistant to inhibitory signals [77]. At the same time, under the influence of IL-2 on NK cells and the induction of cytotoxic activity, no involvement of

another MAPK-pathway, p38, was shown [90]. There was also no evidence of p38 involvement in the induction of NK cell cytotoxicity and in the absence of IL-2 [38]. In addition to VAV-1, the JNK kinase is also required for the realization of the cytotoxic function of NK cells. Violation of the phosphorylation of JNK leads to a violation of the polarization of microtubules and to the violation of immunological synapse formation [38]. In the *in vitro* model, the encephalitis virus (Japanese encephalitis virus) stimulates the shedding of sHLA-E from endothelial cells after infection. The cultivation of NK cells in the presence of media containing sHLA-E resulted in a decrease in the proliferation of NK cells and a decrease in the phosphorylation of ERK1/2 [63].

Thus, when activating NK cells by IL-15 and other cytokines whose receptors contain a common γ-chain (IL-2, IL-7) along the JAK-STAT pathway, the proteins (STAT1, STAT4 and STAT5) intensify the activation, whereas the other proteins (STAT3, STAT6, CIS and SOCS1-7) constrain it. In addition to the JAK-STAT pathway, IL-15 and similar cytokines (IL-2, IL-7) can transmit signal to the cell via signaling pathways associated with the final activation of ERK, JNK and Vav-1 kinases.

NK cells intracellular signaling pathways activation by the IL-12 cytokine

Another cytokine that affects the activation of NK cells is IL-12. In experiments on NK cell lines it was shown that IL-12 induces the synthesis of perforin mRNA [87], stimulates tyrosine phosphorylation in STAT4 proteins [85], but to a lesser extent than IL-2 or IFNγ [85]. In healthy donors, IL-12 causes an increase of STAT4 phosphorylated form in the NK cells [31, 85]. It was shown that the activated form of STAT4 binds to the promoter region of the perforin gene, stimulating its transcription [87]. In healthy donors, IL-12 causes an increase in CD107a expression by NK cells and an increase in the production of IFNγ [31]. After activation of NK cells by IL-12 in patients after liver transplantation, a decrease of phosphorylation of STAT4, which correlates with reduced cytotoxic NK cell function in these patients, is observed [31]. Thus, it can be concluded that STAT4 is involved in the regulation of IL-12-induced cytotoxicity of NK cells [27]. After stimulation of NK cells with IL-12, participation of STAT4 in the induction of expression of T-bet and IL-10 was demonstrated [27]. Currently, for NK cells, by analogy with T lymphocytes, the possibility of antigen-independent and antigen-dependent differentiation and the formation of memory-like NK cells or preactivated NK cells, is discussed [23]. Antigen-dependent differentiation means the formation of a pool of memory-like NK cells as a result of a virus (shown for cytomegalovirus) [35] or as a result of hapten exposure (in mouse experiments) [23]. The antigen-independent differentiation of memory-like NK cells, implies formation of a pool of these cells under the influence of cytokines (IL-12, IL-15 and IL-18) [23, 35]. In a murine model, it was demonstrated that NK cells defective in the gene *IL-12R* fail to form memory-like NK cells after MCMV infection [71]. For NK cells deficient for the gene *IL-12R*, a reduced expression of pSTAT4 is observed [71]. These results demonstrate the necessity of IL-12 stimulation and participation of JAK-STAT cascade in formation of memory-like NK cells, in a murine model [71]. Thus, IL-12 exerts an activating effect on NK cells predominantly with the participation of the JAK-STAT signaling pathway.

NK cells intracellular signaling pathways activation by the TGF-β cytokine

Unlike the cytokines described above, which activate NK cells, TGF-β is a cytokine with immunosuppressive properties. The antagonistic relationship between IL-15 and TGF-β is supported, for example, by the fact that TGF-β inhibits the mTOR kinase activity, the activation of which is induced by IL-15 [82]. It was shown that stimulation of NK cells through CD16 causes an increase in the production of IFNγ by NK cells. After incubation of the NK cells of the NK-92 line in the presence of TGF-β, the production of IFNγ as well as TNFα by the NK cells was reduced [76]. NK cells of the peripheral blood, as well as linear NK cells were used to show that incubation with TGF-β does not lead to an increase in phosphorylated form and STAT4 or ERK activation, but induces the SMAD3 protein phosphorylation [76]. Using the NK-92 cell line it was shown that under the action of TGF-β, NK cells phosphorylation of SMAD2 occurs [88]. Later it was found that NK cells have reduced cytotoxic activity in acute lymphoblastic B cell leukaemia, and inhibition of NK cell function is caused by TGF-β, initiating the signaling pathway of SMAD2/3 proteins [57]. In mice, it was shown that NK cells recovered from a solid tumour and probably exposed to TGF-β from the tumour cells, contained an increased amount of phosphorylated forms of SMAD2/3 [82]. SMAD3 phosphorylation caused decreased production of IFNγ by NK cells and significant inhibition of expression of a gene encoding the transcription factor T-bet [76]. Prolonged incubation of NK cells in the presence of TGF-β resulted in a decrease in their ability to realize antibody-dependent cellular cytotoxicity [76]. This effect was associated with a decreased production of granzyme A and granzyme B due to activation of SMAD3 [76]. Song et al. demonstrated that NK cells after exposure to TGF-β reduced expression of NKG2D receptor [67]. Both NK-92MI cell culture and primary NK cells showed that their incubation in the presence of IL-2 and IL-18 resulted in an increase in NKG2D receptor expression, which was reduced as a result of the previous exposure to TGF-β [67]. Recovery of the NKG2D cy-

totoxic receptor expression by NK cells is associated with the activation of IL-2 and IL-18 signaling pathway related to kinase JNK, but not kinase ERK [67]. In another study, it was also shown that TGF-β causes a decrease in NKG2D expression. Blocking kinases MAPK, ERK and p38 does not lead to restoration of expression of the NKG2D receptor [37]. The results described confirm the specificity of SMAD3 participation in the transmission of an inhibitory signal from TGF-β. Protein SMAD4 regulates signal transduction from TGF-β in NK cell, contributing to the conservation of NK cell expression of TRAIL and inhibiting cell functions via the receptor TGF-βR1 [12]. In a study using mice as model animals, it was found that NK cells defective in the SMAD4 protein exhibit reduced production of IFNγ after stimulation with IL-12 and IL-18 cytokines, as well as reduced cytotoxicity towards target cells [12]. NK cells defective in the SMAD4 protein are also characterized by a reduced expression of the activation receptors CD226 (DNAM-1) and increased expression of the inhibitory receptors TIGIT (WUCAM) [12], high expression of which is characteristic for all ILC1 [12]. Thus, TGF-β suppresses the function of NK cells by inhibiting mTOR and activating SMAD2/3. The absence of inhibition or restoration of the function of NK cells is possible, both through the activation of SMAD-cascade proteins, for example, SMAD4, and through the possible activation of alternative signaling pathways.

Thus, cytokines IL-2, IL-7, IL-15, IL-12 contribute to the differentiation and activation of NK cells using intracellular signaling pathways involving the activation of STAT1, STAT4 and STAT5 and activation of ERK, JNK and Vav-1. Cytokine TGF-β has an immunosuppressive effect on the cytotoxic function of NK cells due to the cascade of intracellular reactions associated with the activation of SMAD 2/3 proteins. In the described cascades, there are proteins which activity blocks the signal from the cytokine (STAT3, SMAD4, etc.) The antagonistic effects of cytokines (IL-15 and TGF-β) on NK cells, as well as the significantly branched cascades of intracellular signaling reactions, determine the complexity of studying NK signaling pathways.

NK cells of the uterus and trophoblast cells: A particular case of the mutual influence of NK cells and microenvironment cells

A special case of tissue-resident NK cells are NK cells of the endometrium and decidua. NK cells significantly change their representation in the uterus, depending on the phase of the menstrual cycle and the onset of pregnancy [44]. Discussion persists on the question of the formation of uterine NK cells. In the decidua, the presence of $CD34^+$ cells were found in humans, which differ from $CD34^+$ peripheral blood and cord blood cells in the spectrum of the mRNA transcription factors. The $CD34^+$ cells of the decidual membrane are characterized by the presence of mRNA transcription factors Nfil3 and Id2, which indicates the possibility of differentiation of NK cells in situ [78]. The possibility of uterine NK cells to differentiate from peripheral blood NK cells and proliferate in situ, is also discussed [9, 44].

Stromal cells and trophoblast cells are the cells of the microenvironment that are able to regulate the formation of the pool of decidual NK cells [44]. According to the literature, decidual stromal cells and trophoblast cells secrete IL-15 [62]. In experiments using mice, IL-15 is shown to be a necessary cytokine for differentiation of uterine NK cells [3]. The IFN-regulatory factor-1 (IRF-1) transcription factor binds to the promoter region of the gene encoding IL-15 [48]. IRF-2 is considered as a functional antagonist of IRF-1 [39]. In experiments with model animals, it is shown that the differentiation of decidual NK cells was disrupted in IRF-1 -/- mice [3]. At the same time, according to other data, the knockout of the gene *IRF-2* had no significant effect on the formation of a pool of decidual NK cells [3, 54]. These results indicate the involvement of IRF-1 in the formation of a pool of decidual NK cells in response to IL-15. In the presence of IL-15, the NK-92 line cells express T-bet and Eomes, and the expression of these transcription factors persists as the IL-15 exposure time increases [49]. Tayade et al. show that at the beginning of pregnancy in murine uterine NK cells, expression of *T-bet* and *Eomes* increased [74]. At the same time, the expression of *Eomes* was significantly superior to the expression of *T-bet* [74]. *T-bet* defective mice did not show disorders of implantation and remodelling of spiral arteries [74]. These results indicate the predominant role of Eomes transcription factor in differentiation of uterine NK cells [74]. At the same time, expression of *T-bet* and *Eomes* increased as the duration of gestation increased, but to a certain level, and then decreased [74]. Park et al. show that the conditioned media of trophoblast cells inhibited the expression of T-bet and Eomes in NK cells previously cultured in the presence of IL-15 [49]. In NK cells, there was also a decrease in the mRNA of perforin and granzyme B when cultivated in the presence of conditioned media of trophoblast cells [49]. A similar effect was shown by Fu et al. on the example of the primary culture of trophoblast cells when cultivated together with decidual NK cells. Trophoblast cells caused reduction of expression of the transcription factor T-bet and Helios in NK cells, which can determine a decrease in IFNγ production and the acquisition of NK cells of the regulatory phenotype [22]. It is likely that at the trophoblast invasion and progressing pregnancy, its inhibitory effect on transcription factors expression of NK cells, associated with terminal stages of differentiation and the acquisition of cytotoxic activity, is enhanced.

In mice, uterine NK cells are shown to be a heterogeneous population consisting of CD49a- cNK cells and CD49a$^+$ NK cells expressing Eomes, as well as CD49a$^+$ Eomes- cells, which are classified as ILC1 [6, 47]. The number of Eomes$^+$CD49a$^+$ NK cells in the uterus decreases during pregnancy, while the number of cNK cells increases [47]. Mice during pregnancy also revealed the presence in the uterus of ILC2 population, expressing GATA-3, and ILC3 population, expressing RORγt, but in much smaller quantities than cNK cells [6]. As in mice, in the human decidua ILC3, expressing RORγt, were detected [19]. At the same time, the number of ILC2, expressing GATA-3, was extremely small [19]. In the analysis of human decidual NK cells, more than 85% of NK cells show the phenotype CD49a$^+$ [21]. It has been determined that CD49a$^+$ NK cells of the decidua containing the transcription factor Eomes express a number of genes (*PTN* (pleiotropine), *OGN* (osteoglycine), *SPP1* (osteopontin), etc.), which products regulate fetal development [21]. The production of HLA-G is characteristic of trophoblast cells. It was shown that the expression of the fetal growth regulating genes in the decidual NK cells was stimulated by extravillous trophoblast cells through the interaction of the NK cell ILT2 receptors and the trophoblast HLA-G [21]. In mice it was shown that defect of *Nfil3*, but not *T-bet*, led to a disruption of the expression of genes *PTN, OGN, SPP1,* which in turn led to weight loss and impaired fetal skeleton [21]. In mice, it was shown that defect of *Nfil3* led to a reduction in the amount cNK cells in the uterus, disruption of spiral arteries remodelling [6] and practically total absence of ILC2, but it did not affect the number ILC1 and ILC3 [19]. According to other data, the knockout of the gene of the transcription factor Nfil3 did not significantly influence the formation of the total pool of decidual NK cells, but led to a disruption in the formation of the placenta [54]. Thus, in connection with the heterogeneity of NK cells population of the decidual membrane, both by surface receptors and transcription factors, their independent differentiation is possible. The transcription factors Eomes, Nfil3, Id2, RORγt and GATA-3 take part in the regulation of differentiation and function of individual populations of decidual NK cells [6, 21].

Cytokine IL-15 activates the proliferation of NK cells and triggers the MAPK signaling pathway, which involves the activation of mitogen-associated protein kinases – ERK and JNK [49]. However, it has been shown that the NK-92 line cells, previously cultured in the presence of IL-15, after exposing the conditioned media of the Sw.71 line trophoblast cells, contained a reduced amount of phosphorylated ERK and JNK compared to the control amount [49]. Also, inhibition of phosphorylation of STAT5 and a decrease in production of perforin, granzyme B and IFNγ by NK cells of the NK-92 line after cultivation in conditioned media of trophoblast cells was shown [49]. When adding collagen synthesized by the primary cell coculture, including trophoblast and decidual stromal cells, to decidual NK cells *in vitro*, a decrease in the number of pSTAT1 and pSTAT4 was observed, which may reflect inhibition of a signaling pathway JAK-STAT [22]. Violation of the blocking of this signaling pathway in decidual NK cells can lead to an increased production of IFNγ by NK cells, which in turn can lead to miscarriage.

The expression and secretion of TGF-β is shown for trophoblast cells [43, 58]. In mice with knockout of the gene ALK5 encoding one of the receptors for TGF-β, the number of uterine NK cells is significantly reduced [50]. ALK5 participates in the transmission of signal from the TGF-β receptor to the SMAD2/3 proteins. Mice with a gene *ALK5* knockout in the uterine NK cells, expressed less genes encoding granzymes (A, B, D, E, F, G), as well as receptor genes to other cytokines [50]. It should be pointed out, that in experiments with mice, one cannot deny the knockout of the same gene in other cells of the microenvironment, which can also make changes to the differentiation process of NK cells of the uterus. For the trophoblast cells, the expression of HLA-E is observed [34]. Using an *in vitro* model with the encephalitis virus engaged to stimulate the shedding of sHLA-E from endothelial cells, it was shown that culturing NK cells in the presence of media containing sHLA-E resulted in a decrease in NK cell proliferation and a decrease in ERK1/2 phosphorylation [63]. In connection with this, the reduction of proliferation of NK cells and the decrease of the activation of NK cells in the decidua along the ERK-dependent pathway due to sHLA-E trophoblast becomes possible. Accordingly, trophoblast cells not only regulate the expression by NK cells of transcription factors of the terminal stages of differentiation, but also inhibit the cytotoxic function of NK cells by suppressing the signaling involving STAT proteins and ERK kinase.

Conclusion

Thus, lymphoid cells of innate immunity (ILC) are now distinguished, which in turn are divided into groups (group 1 ILC, group 2 ILC, group 3 ILC) based on the expression of certain receptors, transcription factors and various functional properties. According to the ILC classification, NK cells are classified as the first group of innate lymphoid cells according to their overall functional characteristics, as well as the participation of the T-bet transcription factor in their differentiation. The complexity and multistage nature of CLP differentiation in mNK are associated with consecutive expression in cells of transcription

factors. Partial regulation by transcription factors of receptor expression to cytokines determines the degree of effect of the cytokine microenvironment on the differentiation of NK cells. Among the cytokines of the microenvironment, IL-2, IL-7, IL-15, IL-12, IL-15 and TGF-β are distinguished, which promote terminal differentiation and regulate the activation of NK cells. The effect of these cytokines has a pleiotropic effect on NK cells and involves the activation of the proteins of the intracellular signaling pathways STAT1, STAT4, STAT5, ERK, JNK, Vav-1, SMAD2, SMAD3 and SMAD4. Depending on the location, the cells of the microenvironment and the spectrum of cytokines secreted by them may differ, which introduces peculiarities in the differentiation of NK cells. For example, it has been found that the population of NK cells of the decidua is heterogeneous in surface receptors and in transcription factors expressed, among which Eomes, Nfil3, Id2, RORγt and GATA-3 are distinguished. Cells of the microenvironment for NK cells in the uterus during pregnancy are stromal decidual cells and trophoblast cells, which to some extent can regulate the heterogeneity of the decidual NK cells. Due to the expression of cytokines and the suppression of signaling involving STAT and ERK proteins, trophoblast cells inhibit the cytotoxic function of NK cells and promote the formation of a pool of regulatory NK cells. The currently known variety of NK cells, depending on the microenvironment in the process of their differentiation, requires further study.

Список литературы / References

1. Ali A.K., Oh J.S., Vivier E., Busslinger M., Lee S.H. NK Cell-specific Gata3 ablation identifies the maturation program required for bone marrow exit and control of proliferation. *J. Immunol., 2016, Vol. 196, no. 4, pp. 1753-1767.*

2. Aliahmad P., de la Torre B., Kaye J. Shared dependence on the DNA-binding factor TOX for the development of lymphoid tissue-inducer cell and NK cell lineages. *Nat. Immunol., 2010, Vol. 11, no. 10, pp. 945-952.*

3. Ashkar A.A., Black G.P., Wei Q., He H., Liang L., Head J.R., Croy B.A. Assessment of requirements for IL-15 and IFN regulatory factors in uterine NK cell differentiation and function during pregnancy. *J. Immunol., 2003, Vol. 171, no. 6, pp. 2937-2944.*

4. Bernink J.H., Peters C.P., Munneke M., te Velde A.A., Meijer S.L., Weijer K., Hreggvidsdottir H.S., Heinsbroek S.E., Legrand N., Buskens C.J., Bemelman W.A., Mjosberg J.M., Spits H. Human type 1 innate lymphoid cells accumulate in inflamed mucosal tissues. *Nat. Immunol., 2013, Vol. 14, no. 3, pp. 221-229.*

5. Billot K., Parizot C., Arrouss I., Mazier D., Debre P., Rogner U.C., Rebollo A. Differential aiolos expression in human hematopoietic subpopulations. *Leuk. Res., 2010, Vol. 34, no. 3, pp. 289-293.*

6. Boulenouar S., Doisne J.M., Sferruzzi-Perri A., Gaynor L.M., Kieckbusch J., Balmas E., Yung H.W., Javadzadeh S., Volmer L., Hawkes D.A., Phillips K., Brady H.J., Fowden A.L., Burton G.J., Moffett A., Colucci F. The residual innate lymphoid cells in NFIL3-deficient mice support suboptimal maternal adaptations to pregnancy. *Front. Immunol., 2016, Vol. 7, p. 43.*

7. Budagian V., Bulanova E., Paus R., Bulfone-Paus S. IL-15/IL-15 receptor biology: a guided tour through an expanding universe. *Cytokine Growth Factor Rev., 2006, Vol. 17, no. 4, pp. 259-280.*

8. Caligiuri M.A., Murray C., Robertson M.J., Wang E., Cochran K., Cameron C., Schow P., Ross M.E., Klumpp T.R., Soiffer R.J., Smith K.A., Ritz J. Selective modulation of human natural killer cells *in vivo* after prolonged infusion of low dose recombinant interleukin 2. *J. Clin. Invest., 1993, Vol. 91, no. 1, pp. 123-132.*

9. Cerdeira A.S., Rajakumar A., Royle C.M., Lo A., Husain Z., Thadhani R.I., Sukhatme V.P., Karumanchi S.A., Kopcow H.D. Conversion of peripheral blood NK cells to a decidual NK-like phenotype by a cocktail of defined factors. *J. Immunol., 2013, Vol. 190, no. 8, pp. 3939-3948.*

10. Chen X., Allan D.S., Krzewski K., Ge B., Kopcow H., Strominger J.L. CD28-stimulated ERK2 phosphorylation is required for polarization of the microtubule organizing center and granules in YTS NK cells. *Proc. Natl. Acad. Sci. USA, 2006, Vol. 103, no. 27, pp. 10346-10351.*

11. Constantinides M.G., McDonald B.D., Verhoef P.A., Bendelac A. A committed precursor to innate lymphoid cells. *Nature, 2014, Vol. 508, no. 7496, pp. 397-401.*

12. Cortez V.S., Ulland T.K., Cervantes-Barragan L., Bando J.K., Robinette M.L., Wang Q., White A.J., Gilfillan S., Cella M., Colonna M. SMAD4 impedes the conversion of NK cells into ILC1-like cells by curtailing non-canonical TGF-beta signaling. *Nat. Immunol., 2017, Vol. 18, no. 9, pp. 995-1003.*

13. Crellin N.K., Trifari S., Kaplan C.D., Satoh-Takayama N., di Santo J.P., Spits H. Regulation of cytokine secretion in human CD127(+) LTi-like innate lymphoid cells by Toll-like receptor 2. *Immunity, 2010, Vol. 33, no. 5, pp. 752-764.*

14. Debierre-Grockiego F. Anti-apoptotic role of STAT5 in haematopoietic cells and in the pathogenesis of malignancies. *Apoptosis, 2004, Vol. 9, no. 6, pp. 717-728.*

15. Delconte R.B., Kolesnik T.B., Dagley L.F., Rautela J., Shi W., Putz E.M., Stannard K., Zhang J.G., Teh C., Firth M., Ushiki T., Andoniou C.E., Degli-Esposti M.A., Sharp P.P., Sanvitale C.E., Infusini G., Liau N.P., Linossi E.M., Burns C.J., Carotta S., Gray D.H., Seillet C., Hutchinson D.S., Belz G.T., Webb A.I., Alexander W.S., Li S.S., Bullock A.N., Babon J.J., Smyth M.J., Nicholson S.E., Huntington N.D. CIS is a potent checkpoint in NK cell-mediated tumor immunity. *Nat. Immunol., 2016, Vol. 17, no. 7, pp. 816-824.*

16. Delconte R.B., Shi W., Sathe P., Ushiki T., Seillet C., Minnich M., Kolesnik T.B., Rankin L.C., Mielke L.A., Zhang J.G., Busslinger M., Smyth M.J., Hutchinson D.S., Nutt S.L., Nicholson S.E., Alexander W.S., Corcoran L.M., Vivier E., Belz G.T., Carotta S., Huntington N.D. The Helix-Loop-Helix protein ID2 governs NK cell fate by tuning their sensitivity to interleukin-15. *Immunity, 2016, Vol. 44, no. 1, pp. 103-115.*

17. Deng Y., Kerdiles Y., Chu J., Yuan S., Wang Y., Chen X., Mao H., Zhang L., Zhang J., Hughes T., Deng Y., Zhang Q., Wang F., Zou X., Liu C.G., Freud A.G., Li X., Caligiuri M.A., Vivier E., Yu J. Transcription factor Foxo1 is a negative regulator of natural killer cell maturation and function. *Immunity, 2015, Vol. 42, no. 3, pp. 457-470.*

18. Diefenbach A., Colonna M., Koyasu S. Development, differentiation, and diversity of innate lymphoid cells. *Immunity, 2014, Vol. 41, no. 3, pp. 354-365.*

19. Doisne J.M., Balmas E., Boulenouar S., Gaynor L.M., Kieckbusch J., Gardner L., Hawkes D.A., Barbara C.F., Sharkey A.M., Brady H.J., Brosens J.J., Moffett A., Colucci F. Composition, development, and function of uterine innate lymphoid cells. *J. Immunol., 2015, Vol. 195, no. 8, pp. 3937-3945.*

20. Erick T.K., Brossay L. Phenotype and functions of conventional and non-conventional NK cells. *Curr. Opin. Immunol., 2016, Vol. 38, pp. 67-74.*

21. Fu B., Zhou Y., Ni X., Tong X., Xu X., Dong Z., Sun R., Tian Z., Wei H. Natural killer cells promote fetal development through the secretion of growth-promoting factors. *Immunity, 2017, Vol. 47, no. 6, pp. 1100-1113 e6.*

22. Fu Q., Sun Y., Tao Y., Piao H., Wang X., Luan X., Du M., Li D. Involvement of the JAK-STAT pathway in collagen regulation of decidual NK cells. *Am. J. Reprod. Immunol., 2017, Vol. 78, no. 6.* doi: 10.1111/aji.12769.

23. Gabrielli S., Ortolani C., del Zotto G., Luchetti F., Canonico B., Buccella F., Artico M., Papa S., Zamai L. The Memories of NK cells: Innate-adaptive immune intrinsic crosstalk. *J. Immunol. Res., 2016, Vol. 2016, 1376595.* doi: 10.1155/2016/1376595.

24. Gordon S.M., Chaix J., Rupp L.J., Wu J., Madera S., Sun J.C., Lindsten T., Reiner S.L. The transcription factors T-bet and Eomes control key checkpoints of natural killer cell maturation. *Immunity, 2012, Vol. 36, no. 1, pp. 55-67.*

25. Gotthardt D., Putz E.M., Grundschober E., Prchal-Murphy M., Straka E., Kudweis P., Heller G., Bago-Horvath Z., Witalisz-Siepracka A., Cumaraswamy A.A., Gunning P.T., Strobl B., Muller M., Moriggl R., Stockmann C., Sexl V. STAT5 Is a key regulator in NK cells and acts as a molecular switch from tumor surveillance to tumor promotion. *Cancer Discov., 2016, Vol. 6, no. 4, pp. 414-429.*

26. Gotthardt D., Putz E.M., Straka E., Kudweis P., Biaggio M., Poli V., Strobl B., Muller M., Sexl V. Loss of STAT3 in murine NK cells enhances NK cell-dependent tumor surveillance. *Blood, 2014, Vol. 124, no. 15, pp. 2370-2379.*

27. Gotthardt D., Sexl V. STATs in NK-Cells: the good, the bad, and the ugly. *Front. Immunol., 2016, Vol. 7, p. 694.*

28. Hazenberg M.D., Spits H. Human innate lymphoid cells. *Blood, 2014, Vol. 124, no. 5, pp. 700-709.*

29. Holmes M.L., Huntington N.D., Thong R.P., Brady J., Hayakawa Y., Andoniou C.E., Fleming P., Shi W., Smyth G.K., Degli-Esposti M.A., Belz G.T., Kallies A., Carotta S., Smyth M.J., Nutt S.L. Peripheral natural killer cell maturation depends on the transcription factor Aiolos. *EMBO J., 2014, Vol. 33, no. 22, pp. 2721-2734.*

30. Hoorweg K., Peters C.P., Cornelissen F., Aparicio-Domingo P., Papazian N., Kazemier G., Mjosberg J.M., Spits H., Cupedo T. Functional differences between human NKp44(-) and NKp44(+) RORC(+) innate lymphoid cells. *Front. Immunol., 2012, Vol. 3, p. 72.*

31. Jamil K.M., Hydes T.J., Cheent K.S., Cassidy S.A., Traherne J.A., Jayaraman J., Trowsdale J., Alexander G.J., Little A.M., McFarlane H., Heneghan M.A., Purbhoo M.A., Khakoo S.I. STAT4-associated natural killer cell tolerance following liver transplantation. *Gut, 2017, Vol. 66, no. 2, pp. 352-361.*

32. Jiang K., Zhong B., Gilvary D.L., Corliss B.C., Vivier E., Hong-Geller E., Wei S., Djeu J.Y. Syk regulation of phosphoinositide 3-kinase-dependent NK cell function. *J. Immunol., 2002, Vol. 168, no. 7, pp. 3155-3164.*

33. Judge C.J., Kostadinova L., Sherman K.E., Butt A.A., Falck-Ytter Y., Funderburg N.T., Landay A.L., Lederman M.M., Sieg S.F., Sandberg J.K., Anthony D.D. CD56^{bright} NK IL-7Ralpha expression negatively associates with HCV level, and IL-7-induced NK function is impaired during HCV and HIV infections. *J. Leukoc. Biol., 2017, Vol. 102, no. 1, pp. 171-184.*

34. King A., Allan D.S., Bowen M., Powis S.J., Joseph S., Verma S., Hiby S.E., McMichael A.J., Loke Y.W., Braud V.M. HLA-E is expressed on trophoblast and interacts with CD94/NKG2 receptors on decidual NK cells. *Eur. J. Immunol., 2000, Vol. 30, no. 6, pp. 1623-1631.*

35. Kovalenko E.I., Streltsova M.A. Adaptive features of natural killer cells, lymphocytes of innate immunity. *Russ. J. Bioorganic Chem., 2016, Vol. 42, no. 6, pp. 590-605.*

36. Kucuk C., Jiang B., Hu X., Zhang W., Chan J.K., Xiao W., Lack N., Alkan C., Williams J.C., Avery K.N., Kavak P., Scuto A., Sen E., Gaulard P., Staudt L., Iqbal J., Zhang W., Cornish A., Gong Q., Yang Q., Sun H., d'Amore F., Leppa S., Liu W., Fu K., de Leval L., McKeithan T., Chan W.C. Activating mutations of STAT5B and STAT3 in lymphomas derived from gammadelta-T or NK cells. *Nat. Commun., 2015, Vol. 6, p. 6025.*

37. Lee J.C., Lee K.M., Ahn Y.O., Suh B., Heo D.S. A possible mechanism of impaired NK cytotoxicity in cancer patients: down-regulation of DAP10 by TGF-beta1. *Tumori, 2011, Vol. 97, no. 3, pp. 350-357.*

38. Li C., Ge B., Nicotra M., Stern J.N., Kopcow H.D., Chen X., Strominger J.L. JNK MAP kinase activation is required for MTOC and granule polarization in NKG2D-mediated NK cell cytotoxicity. *Proc. Natl. Acad. Sci. USA, 2008, Vol. 105, no. 8, pp. 3017-3022.*

39. Lohoff M., Duncan G.S., Ferrick D., Mittrucker H.W., Bischof S., Prechtl S., Rollinghoff M., Schmitt E., Pahl A., Mak T.W. Deficiency in the transcription factor interferon regulatory factor (IRF)-2 leads to severely compromised development of natural killer and T helper type 1 cells. *J. Exp. Med., 2000, Vol. 192, no. 3, pp. 325-336.*

40. Male V., Brady H.J. Transcriptional control of NK cell differentiation and function. *Curr. Top Microbiol. Immunol., 2014, Vol. 381, pp. 173-187.*

41. Male V., Nisoli I., Kostrzewski T., Allan D.S., Carlyle J.R., Lord G.M., Wack A., Brady H.J. The transcription factor E4bp4/Nfil3 controls commitment to the NK lineage and directly regulates Eomes and Id2 expression. *J. Exp. Med., 2014, Vol. 211, no. 4, pp. 635-642.*

42. Mao Y., van Hoef V., Zhang X., Wennerberg E., Lorent J., Witt K., Masvidal L., Liang S., Murray S., Larsson O., Kiessling R., Lundqvist A. IL-15 activates mTOR and primes stress-activated gene expression leading to prolonged antitumor capacity of NK cells. *Blood, 2016, Vol. 128, no. 11, pp. 1475-1489.*

43. Matsumoto J., Kawana K., Nagamatsu T., Schust D.J., Fujii T., Sato H., Hyodo H., Yasugi T., Kozuma S., Taketani Y. Expression of surface CD1d in the extravillous trophoblast cells of early gestational placenta is downregulated in a manner dependent on trophoblast differentiation. *Biochem. Biophys. Res. Commun., 2008, Vol. 371, no. 2, pp. 236-241.*

44. Mikhailova V.A., Belyakova K.L., Selkov S.A., Sokolov D.I. Peculiarities of NK cells differentiation: $CD56^{dim}$ and $CD56^{bright}$ NK cells at pregnancy and in non-pregnant state. *Meditsinskaya immunologiya = Medical Immunology (Russia), 2017, Vol. 19, no. 1, pp. 19-26.* doi: 10.15789/1563-0625-2017-1-19-26.

45. Mikhaylova V.A., Selkov S.A., Sokolov D.I. Phenotypic and functional characteristics of NK cells in pregnancy. *Scientific and Practical Journal "Obstetrics and Gynecology", 2011, Vol. 5, pp. 4-9.*

46. Mishra A., Sullivan L., Caligiuri M.A. Molecular pathways: interleukin-15 signaling in health and in cancer. *Clin. Cancer Res., 2014, Vol. 20, no. 8, pp. 2044-2050.*

47. Montaldo E., Vacca P., Chiossone L., Croxatto D., Loiacono F., Martini S., Ferrero S., Walzer T., Moretta L., Mingari M.C. Unique eomes(+) NK cell subsets are present in uterus and decidua during early pregnancy. *Front. Immunol., 2015, Vol. 6, p. 646.*

48. Ogasawara K., Hida S., Azimi N., Tagaya Y., Sato T., Yokochi-Fukuda T., Waldmann T.A., Taniguchi T., Taki S. Requirement for IRF-1 in the microenvironment supporting development of natural killer cells. *Nature, 1998, Vol. 391, no. 6668, pp. 700-703.*

49. Park S.Y., Yun S., Ryu B.J., Han A.R., Lee S.K. Trophoblasts regulate natural killer cells via control of interleukin-15 receptor signaling. *Am. J. Reprod. Immunol., 2017, Vol. 78, no. 2.* doi: 10.1111/aji.12628.

50. Peng J., Monsivais D., You R., Zhong H., Pangas S.A., Matzuk M.M. Uterine activin receptor-like kinase 5 is crucial for blastocyst implantation and placental development. *Proc. Natl. Acad. Sci. USA, 2015, Vol. 112, no. 36, E5098-107.* doi :10.1073/pnas.1514498112.

51. Pinho M.J., Marques C.J., Carvalho F., Punzel M., Sousa M., Barros A. Genetic regulation on *ex vivo* differentiated natural killer cells from human umbilical cord blood $CD34^{+}$ cells. *J. Recept. Signal Transduct Res., 2012, Vol. 32, no. 5, pp. 238-249.*

52. Ramirez K., Chandler K.J., Spaulding C., Zandi S., Sigvardsson M., Graves B.J., Kee B.L. Gene deregulation and chronic activation in natural killer cells deficient in the transcription factor ETS1. *Immunity, 2012, Vol. 36, no. 6, pp. 921-932.*

53. Rautela J., Huntington N.D. IL-15 signaling in NK cell cancer immunotherapy. *Curr. Opin. Immunol., 2017, Vol. 44, pp. 1-6.*

54. Redhead M.L., Portilho N.A., Felker A.M., Mohammad S., Mara D.L., Croy B.A. The transcription factor NFIL3 is essential for normal placental and embryonic development but not for uterine natural killer (UNK) cell differentiation in mice. *Biol. Reprod., 2016, Vol. 94, no. 5, p. 101.*

55. Robbins S.H., Tessmer M.S., van Kaer L., Brossay L. Direct effects of T-bet and MHC class I expression, but not STAT1, on peripheral NK cell maturation. *Eur. J. Immunol., 2005, Vol. 35, no. 3, pp. 757-765.*

56. Robinette M.L., Fuchs A., Cortez V.S., Lee J.S., Wang Y., Durum S.K., Gilfillan S., Colonna M. Immunological Genome C. Transcriptional programs define molecular characteristics of innate lymphoid cell classes and subsets. *Nat. Immunol., 2015, Vol. 16, no. 3, pp. 306-317.*

57. Rouce R.H., Shaim H., Sekine T., Weber G., Ballard B., Ku S., Barese C., Murali V., Wu M.F., Liu H., Shpall E.J., Bollard C.M., Rabin K.R., Rezvani K. The TGF-beta/SMAD pathway is an important mechanism for NK cell immune evasion in childhood B-acute lymphoblastic leukemia. *Leukemia, 2016, Vol. 30, no. 4, pp. 800-811.*

58. Schilling B., Yeh J. Transforming growth factor-beta(1), -beta(2), -beta(3) and their type I and II receptors in human term placenta. *Gynecol. Obstet. Invest., 2000, Vol. 50, no. 1, pp. 19-23.*

59. Scoville S.D., Freud A.G., Caligiuri M.A. Modeling human natural killer cell development in the era of innate lymphoid cells. *Front. Immunol., 2017, Vol. 8, p. 360.*

60. Seillet C., Belz G.T., Huntington N.D. Development, homeostasis, and heterogeneity of NK cells and ILC1. *Curr. Top Microbiol. Immunol., 2016, Vol. 395, pp. 37-61.*

61. Seillet C., Huntington N.D., Gangatirkar P., Axelsson E., Minnich M., Brady H.J., Busslinger M., Smyth M.J., Belz G.T., Carotta S. Differential requirement for Nfil3 during NK cell development. *J. Immunol., 2014, Vol. 192, no. 6, pp. 2667-2676.*

62. Sharma S., Godbole G., Modi D. Decidual control of trophoblast invasion. *Am. J. Reprod. Immunol., 2016, Vol. 75, no. 3, pp. 341-350.*

63. Shwetank, Date O.S., Carbone E., Manjunath R. Inhibition of ERK and proliferation in NK cell lines by soluble HLA-E released from Japanese encephalitis virus infected cells. *Immunol. Lett., 2014, Vol. 162, no. 1, Pt A, pp. 94-100.*

64. Sim G.C., Wu S., Jin L., Hwu P., Radvanyi L.G. Defective STAT1 activation associated with impaired IFN-gamma production in NK and T lymphocytes from metastatic melanoma patients treated with IL-2. *Oncotarget, 2016, Vol. 7, no. 24, pp. 36074-36091.*

65. Simonetta F., Pradier A., Bosshard C., Masouridi-Levrat S., Chalandon Y., Roosnek E. NK cell functional impairment after allogeneic hematopoietic stem cell transplantation is associated with reduced levels of T-bet and eomesodermin. *J. Immunol., 2015, Vol. 195, no. 10, pp. 4712-4720.*

66. Simoni Y., Newell E.W. Dissecting human ILC heterogeneity: more than just three subsets. *Immunology, 2018, Vol. 153, no. 3, pp. 297-303.*

67. Song H., Hur D.Y., Kim K.E., Park H., Kim T., Kim C.W., Bang S., Cho D.H. IL-2/IL-18 prevent the down-modulation of NKG2D by TGF-beta in NK cells via the c-Jun N-terminal kinase (JNK) pathway. *Cell Immunol., 2006, Vol. 242, no. 1, pp. 39-45.*

68. Spits H., Artis D., Colonna M., Diefenbach A., di Santo J.P., Eberl G., Koyasu S., Locksley R.M., McKenzie A.N., Mebius R.E., Powrie F., Vivier E. Innate lymphoid cells – a proposal for uniform nomenclature. *Nat. Rev. Immunol., 2013, Vol. 13, no. 2, pp. 145-149.*

69. Spits H., Bernink J.H., Lanier L. NK cells and type 1 innate lymphoid cells: partners in host defense. *Nat. Immunol., 2016, Vol. 17, no. 7, pp. 758-764.*

70. Spits H., Cupedo T. Innate lymphoid cells: emerging insights in development, lineage relationships, and function. *Annu Rev. Immunol., 2012, Vol. 30, pp. 647-675.*

71. Sun J.C., Madera S., Bezman N.A., Beilke J.N., Kaplan M.H., Lanier L.L. Proinflammatory cytokine signaling required for the generation of natural killer cell memory. *J. Exp. Med., 2012, Vol. 209, no. 5, pp. 947-954.*

72. Tait Wojno E.D., Artis D. Innate lymphoid cells: balancing immunity, inflammation, and tissue repair in the intestine. *Cell Host Microbe, 2012, Vol. 12, no. 4, pp. 445-457.*

73. Tanriver Y., Diefenbach A. Transcription factors controlling development and function of innate lymphoid cells. *Int. Immunol., 2014, Vol. 26, no. 3, pp. 119-128.*

74. Tayade C., Fang Y., Black G.P., V A.P., Jr., Erlebacher A., Croy B.A. Differential transcription of Eomes and T-bet during maturation of mouse uterine natural killer cells. *J. Leukoc. Biol., 2005, Vol. 78, no. 6, pp. 1347-1355.*

75. Townsend M.J., Weinmann A.S., Matsuda J.L., Salomon R., Farnham P.J., Biron C.A., Gapin L., Glimcher L.H. T-bet regulates the terminal maturation and homeostasis of NK and Valpha14i NKT cells. *Immunity, 2004, Vol. 20, no. 4, pp. 477-494.*

76. Trotta R., Dal Col J., Yu J., Ciarlariello D., Thomas B., Zhang X., Allard J., 2nd, Wei M., Mao H., Byrd J.C., Perrotti D., Caligiuri M.A. TGF-beta utilizes SMAD3 to inhibit CD16-mediated IFN-gamma production and antibody-dependent cellular cytotoxicity in human NK cells. *J. Immunol., 2008, Vol. 181, no. 6, pp. 3784-3792.*

77. Urlaub D., Bhat R., Messmer B., Watzl C. Co-activation of cultured human natural killer cells: enhanced function and decreased inhibition. *J. Toxicol. Environ Health A, 2016, Vol. 79, no. 22-23, pp. 1078-1084.*

78. Vacca P., Vitale C., Montaldo E., Conte R., Cantoni C., Fulcheri E., Darretta V., Moretta L., Mingari M.C. $CD34^+$ hematopoietic precursors are present in human decidua and differentiate into natural killer cells upon interaction with stromal cells. *Proc. Natl. Acad. Sci. USA, 2011, Vol. 108, no. 6, pp. 2402-2407.*

79. van de Walle I., Dolens A.C., Durinck K., de Mulder K., van Loocke W., Damle S., Waegemans E., de Medts J., Velghe I., de Smedt M., Vandekerckhove B., Kerre T., Plum J., Leclercq G., Rothenberg E.V., van Vlierberghe P., Speleman F., Taghon T. GATA3 induces human T-cell commitment by restraining Notch activity and repressing NK-cell fate. *Nat. Commun., 2016, Vol. 7, 11171.* doi: 10.1038/ncomms11171.

80. van Helden M.J., Goossens S., Daussy C., Mathieu A.L., Faure F., Marcais A., Vandamme N., Farla N., Mayol K., Viel S., Degouve S., Debien E., Seuntjens E., Conidi A., Chaix J., Mangeot P., de Bernard S., Buffat L., Haigh J.J., Huylebroeck D., Lambrecht B.N., Berx G., Walzer T. Terminal NK cell maturation is controlled by concerted actions of T-bet and Zeb2 and is essential for melanoma rejection. *J. Exp. Med., 2015, Vol. 212, no. 12, pp. 2015-2025.*

81. Vargas-Hernandez A., Mace E.M., Zimmerman O., Zerbe C.S., Freeman A.F., Rosenzweig S., Leiding J.W., Torgerson T., Altman M.C., Schussler E., Cunningham-Rundles C., Chinn I.K., Carisey A.F., Hanson I.C., Rider N.L., Holland S.M., Orange J.S., Forbes L.R. Ruxolitinib partially reverses functional NK cell deficiency in patients with STAT1 gain-of-function mutations. *J. Allergy Clin. Immunol., 2018, Vol. 141, no. 6, pp. 2142-2155.*

82. Viel S., Marcais A., Guimaraes F.S., Loftus R., Rabilloud J., Grau M., Degouve S., Djebali S., Sanlaville A., Charrier E., Bienvenu J., Marie J.C., Caux C., Marvel J., Town L., Huntington N.D., Bartholin L., Finlay D., Smyth M.J., Walzer T. TGF-beta inhibits the activation and functions of NK cells by repressing the mTOR pathway. *Sci. Signal., 2016, Vol. 9, no. 415, ra19.* doi: 10.1126/scisignal.aad1884.

83. Vong Q.P., Leung W.H., Houston J., Li Y., Rooney B., Holladay M., Oostendorp R.A., Leung W. TOX2 regulates human natural killer cell development by controlling T-BET expression. *Blood, 2014, Vol. 124, no. 26, pp. 3905-3913.*

84. Wagner J.A., Rosario M., Romee R., Berrien-Elliott M.M., Schneider S.E., Leong J.W., Sullivan R.P., Jewell B.A., Becker-Hapak M., Schappe T., Abdel-Latif S., Ireland A.R., Jaishankar D., King J.A., Vij R., Clement D., Goodridge J., Malmberg K.J., Wong H.C., Fehniger T.A. $CD56^{bright}$ NK cells exhibit potent antitumor responses following IL-15 priming. *J. Clin. Invest., 2017, Vol. 127, no. 11, pp. 4042-4058.*

85. Wang K.S., Ritz J., Frank D.A. IL-2 induces STAT4 activation in primary NK cells and NK cell lines, but not in T cells. *J. Immunol., 1999, Vol. 162, no. 1, pp. 299-304.*

86. Wang S., Xia P., Huang G., Zhu P., Liu J., Ye B., Du Y., Fan Z. FoxO1-mediated autophagy is required for NK cell development and innate immunity. *Nat. Commun., 2016, Vol. 7, 11023.* doi: 10.1038/ncomms11023.

87. Yamamoto K., Shibata F., Miyasaka N., Miura O. The human perforin gene is a direct target of STAT4 activated by IL-12 in NK cells. *Biochem. Biophys. Res. Commun., 2002, Vol. 297, no. 5, pp. 1245-1252.*

88. Yang B., Liu H., Shi W., Wang Z., Sun S., Zhang G., Hu Y., Liu T., Jiao S. Blocking transforming growth factor-beta signaling pathway augments antitumor effect of adoptive NK-92 cell therapy. *Int. Immunopharmacol., 2013, Vol. 17, no. 2, pp. 198-204.*

89. Yokota Y., Mansouri A., Mori S., Sugawara S., Adachi S., Nishikawa S., Gruss P. Development of peripheral lymphoid organs and natural killer cells depends on the helix-loop-helix inhibitor Id2. *Nature, 1999, Vol. 397, no. 6721, pp. 702-706.*

90. Yu T.K., Caudell E.G., Smid C., Grimm E.A. IL-2 activation of NK cells: involvement of MKK1/2/ERK but not p38 kinase pathway. *J. Immunol., 2000, Vol. 164, no. 12, pp. 6244-6251.*

Авторы:

Михайлова В.А. *— к.б.н., старший научный сотрудник лаборатории межклеточных взаимодействий, отдел иммунологии и межклеточных взаимодействий ФГБНУ «Научно-исследовательский институт акушерства, гинекологии и репродуктологии имени Д.О. Отта»; кафедра иммунологии ГБОУ ВПО «Первый Санкт-Петербургский государственный медицинский университет имени академика И.П. Павлова» Министерства здравоохранения РФ, Санкт-Петербург, Россия*

Баженов Д.О. *— младший научный сотрудник лаборатории межклеточных взаимодействий, отдел иммунологии и межклеточных взаимодействий ФГБНУ «Научно-исследовательский институт акушерства, гинекологии и репродуктологии имени Д.О. Отта», Санкт-Петербург, Россия*

Authors:

Mikhailova V.A., *PhD (Biology), Senior Research Associate, Laboratory of Intercellular Interactions, Department of Immunology and Intercellular Interactions, D. Ott Research Institute of Obstetrics, Gynecology and Reproductology; Department of Immunology, First St. Petersburg State I. Pavlov Medical University, St. Petersburg, Russian Federation*

Bazhenov D.O., *Junior Research Associate, Laboratory of Intercellular Interactions, Department of Immunology and Intercellular Interactions, D. Ott Research Institute of Obstetrics, Gynecology and Reproductology, St. Petersburg, Russian Federation*

Белякова К.Л. *– младший научный сотрудник лаборатории межклеточных взаимодействий, отдел иммунологии и межклеточных взаимодействий ФГБНУ «Научно-исследовательский институт акушерства, гинекологии и репродуктологии имени Д.О. Отта», Санкт-Петербург, Россия*

Selkov S.A., *PhD, MD (Medicine), Professor, Honored Science Worker, Head, Department of Immunology and Intercellular Interactions, D. Ott Research Institute of Obstetrics, Gynecology and Reproductology, St. Petersburg, Russian Federation*

Сельков С.А. *– д.м.н., профессор, заслуженный деятель науки, заведующий отделом иммунологии и межклеточных взаимодействий ФГБНУ «Научно-исследовательский институт акушерства, гинекологии и репродуктологии имени Д.О. Отта», Санкт-Петербург, Россия*

Sokolov D.I., *PhD, MD (Biology), Head, Laboratory of Intercellular Interactions, Department of Immunology and Intercellular Interactions, D. Ott Research Institute of Obstetrics, Gynecology and Reproductology; Department of Immunology, First St. Petersburg State I. Pavlov Medical University; Department of Immunology, Institute of Experimental Medicine, St. Petersburg, Russian Federation*

Соколов Д.И. *– д.б.н., заведующий лабораторией межклеточных взаимодействий, отдел иммунологии и межклеточных взаимодействий ФГБНУ «Научно-исследовательский институт акушерства, гинекологии и репродуктологии имени Д.О. Отта»; кафедра иммунологии ГБОУ ВПО «Первый Санкт-Петербургский государственный медицинский университет имени академика И.П. Павлова» Министерства здравоохранения РФ; отдел иммунологии ФГБНУ «Институт экспериментальной медицины», Санкт-Петербург, Россия*

Поступила 30.03.2018
Принята к печати 05.04.2018

Received 30.03.2018
Accepted 05.04.2018