 DOI: https://doi.org/10.5114/ceji.2021.106998

Expression of *ORAI1* and *STIM1* genes in blood of patients with pulmonary tuberculosis

OKSANA KOLESOVA, KSENIJA KRAMICA, ALEKSANDRS KOLESOVS, JELENA EGLITE

Riga Stradins University, Latvia

Abstract

This study aimed to detect the expression level of ORAI1 and STIM1 genes in blood of patients with bilateral pulmonary tuberculosis (TB) in comparison with the control group. Both genes encode proteins providing store-operated Ca^{2+} entry (SOCE) into the cells, including immune cells, to activate transcriptional factors for producing cytokines and inflammation-restricting proteins. The study included 45 patients with confirmed TB, aged 20 to 86, and 35 volunteers, aged from 21 to 73, without active TB infection. The expression of ORAI1 and STIM1 genes in blood was performed by real-time quantitative reverse transcription polymerase chain reaction (RT-qPCR). Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) was used as the referent gene. Inflammation was assessed by levels of interferon γ (IFN-γ) and interleukin 18 (IL-18) in serum (ELISA method). The results showed lower expression of ORAI1 in blood and higher levels of IFN-γ and IL-18 in serum of TB patients than that of the control group and no differences in expression of the STIM1 gene. It indicates some impairment in the SOCE mechanism of immune cells, which is associated with TB.

Key words: *cytokines, tuberculosis, SOCE, mRNA ORAI1, mRNA STIM1.*

(Cent Eur J Immunol 2021; 46 (2): 275-282)

Introduction

ORAI1 and *STIM1* genes are essential for the store-operated Ca^{2+} entry (SOCE) mechanism used by immune cells to increase intracellular Ca^{2+} concentrations [1]. Expression of these genes in blood of patients with tuberculosis (TB) became the focus of our exploration. We expected some impairment in this specific regulatory mechanism of Ca^{2+} entry in patients with TB.

Tuberculosis is an infection characterized by the persistence of mycobacteria in macrophages and dendritic cells due to imperfect phagocytosis. The local secretion of interferon γ (IFN-γ), produced by type 1 T helper cells, and tumor necrosis factor α (TNF-α), primarily produced by macrophages, are the main factors enhancing the maturation of phagolysosomes, production of reactive oxygen (ROS), nitrogen species, and antimicrobial peptides in macrophages [2]. Activation of immune cells, production of cytokines and inflammation-restricting proteins (Fas-ligand, Foxp3), and production of ROS in phagosomes of phagocytes occur after an increase of intracellular Ca^{2+} concentration [3, 4]. Many studies [5-9] indicate that mycobacteria use different strategies to manipulate Ca^{2+} signaling in macrophages and dendritic cells to increase their lifetime and escape from the immune response.

There are two main channels providing Ca^{2+} influx into the immune cells: Ca^{2+} release-activated Ca^{2+} (CRAC) channels, which are activated by decreasing intracellular Ca^{2+} concentration, and voltage-gated Ca^{2+} channels (VGCC), which are opened by depolarization of the cytoplasmic membrane [5, 10]. The immune cells predominantly use CRAC channels of SOCE [1]. The prototypical CRAC channel is formed by ORAI1 protein (encoded by the *ORAI1* gene) located on the cytoplasmic membrane. Its activation is provided by ligation of stromal interaction molecule 1 (STIM1, encoded by the *STIM1* gene) located on the membrane of the endoplasmic reticulum [10].

The importance of *STIM1* and *ORAI1* genes in TB was demonstrated in an experimental model in TB infected mice [3]. A defect of the *STIM1* gene led to the death of mice due to pulmonary hyper-inflammation and loss of respiratory function. It suggests that a dysfunction of SOCE may contribute to imperfect mycobacterial phagocytosis and provide a basis for mycobacterial persistence. Simultaneously, there are no data about the expression of *ORAI1* and *STIM1* genes in humans with TB. Detection of the expression level of *ORAI1* and *STIM1* genes in blood despite their non-specificity for TB can provide additional information for a TB pathogenesis, prognosis, and monitoring of TB treatment.

This study aimed to detect the level of expression of *ORAI1* and *STIM1* genes in blood of TB patients at the beginning of anti-TB treatment in association with immunological status, assessed by levels of IFN-γ and interleukin 18 (IL-18) in serum, and to compare the expression of

Correspondence: Oksana Kolesova, PhD, Riga Stradins University, 5 Ratsupites St., Riga, Latvia, LV1067, e-mail: oksana-kolesova@inbox.lv
Submitted: 7.03.2020, Accepted: 17.07.2020

This is an Open Access article distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International (CC BY-NC-SA 4.0). License (http://creativecommons.org/licenses/by-nc-sa/4.0/)

ORAI1 and *STIM1* with a healthy control group. IL-18 was selected for the assessment of the immunological status as a pro-inflammatory cytokine, produced predominantly by activated monocytes/macrophages and having broad immunological effects [11], including an increase of production of IFN-γ by T cells and enhancing the protective Th1 immune response against mycobacteria. Simultaneously, increasing activity of IL-18 induces activation of nuclear factor-kB and expression of Fas ligands, which can lead to tissue destruction [11, 12]. An increasing level of IL-18 in serum also has a negative predictive value for HIV-associated TB [13].

Material and methods

The permission of the Ethics Committee of Riga East University Hospital (No. 9-A/17) was obtained for the study. The study included 45 patients, aged 20 to 86 years (mean age = 49.7 years, SD = 16.0, 33% females), with confirmed TB. The patients underwent observation and treatment from October 2017 to December 2019 in the Lung Disease and Tuberculosis Ward of Daugavpils Regional Hospital (Latvia). Inclusion criteria were: age of 18 and older, bacteriologically confirmed pulmonary TB, and drug-sensitive TB. Exclusion criteria were: age < 18, exclusively extrapulmonary TB, pulmonary TB with concurrent extrapulmonary TB involvement, pregnancy, imprisonment, mental disorders, and HIV-positive status. All patients had bilateral drug-sensitive tuberculous pneumonia and used first-line anti-tuberculous drugs according to the standard regimen.

The control group included 35 volunteers, aged from 21 to 73 years (mean age = 36.8 years, SD = 16.0, 30% females), without active TB and human immunodeficiency virus (HIV) infection. For the control group, the inclusion criteria were age of 18 and older, self-reported physical health, and no ongoing infection.

For analysis of *ORAI1* and *STIM1* expression, 1 ml of peripheral blood with EDTA was collected and stored at –20ºC before detection. RNA was extracted from stored blood samples using the innuPREP Blood RNA Life Science Kits & Assays (Analytik Jena Company, Germany) according to the manufacturer's procedures. The quality and quantity of extracted RNA were detected by spectrophotometry using the Nanofotometr NF80 (Implen GmbH, München, Germany). The concentration of RNA was 40 μg/ml per reaction. For reverse transcription-PCR (RT-qPCR), the Revert First-Strand cDNA synthesis system with an oligo dT primer (QuantiTect Reverse Transcription, an oligo dT primer Invitrogen, Germany) was used following the manufacturer's instructions. The first-strand cDNA was diluted 1 : 20 with distilled water with the following used as a template (K1+) in RT-qPCR analysis. Specific primers for human *STIM1* (Hs STIM1 FAM_1, QF00208159) and *ORAI1* (Hs ORAI 1 FAM_1, QF00163611) were detected by one-step qRT-PCR using sequence-specific probes for gene expression analysis (QuantiFast Probe Assay, Invitrogen, Germany). RT-qPCR was performed in a DTlite Cycler (DNA-Technology, Russia) by the fast real-time PCR System using the following amplification conditions: 5 min of initial denaturation at 95˚C, then 45 cycles of 95˚C for 30 s, 60˚C for 30 s. The specificity of RT-qPCR products was confirmed by the analysis of a melting curve. Absolute quantification of *STIM1* and *ORAI1* gene expression was detected relative to a standard curve, automatically created by serial dilution of the reference gene glyceraldehyde-3-phosphate dehydrogenase (*GAPDH*). For amplification, the following primers (Invitrogen by Thermo Fischer Scientific) and reference gene (Bioneer Corporation, Republic of Korea) were used: *ORAI1*, forward: 5′-CGTATCTAGAATGCATCCGGAGCC-3′, reverse: 5′-CAGCCACTATGCCTAGGTCGACTAGC-3′; *STIM1*, forward: 5′-CCTCGGTACCATCCATGTTGTAGCA-3′, reverse: 5′-GCGAAAGCTTACGCTAAAATGGTGTCT-3′; *GAPDH*, forward: 5′-CCACTCCTCCACCTTTGAC-3′, reverse: 5′-ACCCTGTTGCTGTAGCCA-3′; Probe, 5′-TTGCCCTCAACGACCACTTTGTC-3′.

Detection of IFN-γ and IL-18 in serum was performed by the sandwich immunoassay method using the commercial kits *IFA-BEST INF-γ* (detection range: 0-1000 pg/ml, sensitivity: 2.0 pg/ml, normal values: < 15 pg/ml) and *IFA-BEST IL-18* (detection range: 0-1000 pg/ml, sensitivity: 2.0 pg/ml, normal values: 90-260 pg/ml) according to the methodology of the manufacturer (Vector-Best, Russia).

Results and discussion

The Shapiro-Wilk test revealed significant deviance from the normal distribution in both groups of participants. Test values varied from 0.19 to 0.91 ($p < 0.01$ to $p < 0.001$). Observed non-normality of distributions led to the use of nonparametric statistics for a comparison of the control and TB groups. The results of the Mann-Whitney *U*-test (Table 1) indicated a significantly higher level of mRNA *ORAI1* in the control group.

Simultaneously, there were no significant differences between groups in the level of mRNA *STIM1*. The level of IFN-γ and IL-18 were higher in the group of TB patients. Figure 1 presents the comparison between groups.

Expression levels of *ORAI1* and *STIM1* genes in blood dominantly reflected their expression in peripheral blood leukocytes, being immune cells. A more expressed *ORAI1* gene in TB patients indicated an impaired SOCE mechanism in these cells.

Taking into account that ORAI1 protein forms a pore in the plasmatic membrane of immune cells for releasing Ca^{2+} ions into cytoplasm, a lower expression level of *ORAI1* in blood can be associated with a lower influx of Ca^{2+} into cells, leading to lower cytokine production. In

Table 1. Comparison of markers in tuberculosis (TB) and control groups

Markers	TB group (*n* = 45) Median (IQR)	Control group (*n* = 35) Median (IQR)	Mann-Whitney *U*-test
mRNA ORAI1 (copies/ml)	55.7 (1.7-520.5)	1.3E+12 (1.1E+9-2.6E+13)	1.0***
mRNA STIM1 (copies/ml)	978.0 (7.4-3230.0)	383.0 (9.2-21200.0)	778.0
IFN-γ (pg/ml)	39.3[a] (17.3-79.2)	0.0[b] (0.0-0.0)	10.0***
IL-18 (pg/ml)	276.9[c] (228.4-397.8)	225.8[b] (208.2-253.1)	208.0*

*IQR – interquartile range, [a] presented for a subsample n = 34, [b] presented for a subsample n = 15, [c] presented for a subsample n = 44, *p < 0.05, ***p < 0.001*

contrast, our findings showed that the production of IFN-γ and IL-18 is not disrupted in patients with TB and is at a higher level than in the control group. As described in a model of TB infected mice [3], impaired SOCE is associated with the reduced expression of inflammation-restricting proteins, leading to hyper-inflammation.

The Spearman correlation coefficients indicated no significant relationships among the markers in both the control and TB groups (Table 2). It indicates the relative independence of the level of expression of genes and the level of inflammation.

Simultaneously, non-pronounced differences in *STIM1* gene expression indicate a low association of its expression with TB. Ca^{2+} influx into immune cells is also provided by other Ca^{2+} channels [14], for example, VGCC [6]. In this case, Ca^{2+} influx through VGCC leads to suppression of the protective immune response [6, 7].

At a more generalized level, observed differences in expression of the *ORAI1* gene have at least two explanations. One the one hand, low expression of the *ORAI1* gene in patients with TB can be a result of mycobacterial infection. Previous studies [15-18] show that mycobacteria can change Ca^{2+} signaling and protect survival in macrophages by regulation of expression of microRNAs (endogenous regulators of gene expression) in TB patients. It is possible that microRNAs block *ORAI1* expression. The relationship between *ORAI1* expression and microRNA specific for TB can be the further direction of the investigation. On the other hand, we have not assessed the dynamics of *ORAI1* expression. A low level of expression can be observed before the infection and affect the development of TB. A prospective study can present the dynamics of *ORAI1* after the treatment. In addition, low expression of *ORAI1* can be a result of genetic polymorphisms of the *ORAI1* gene [19, 20].

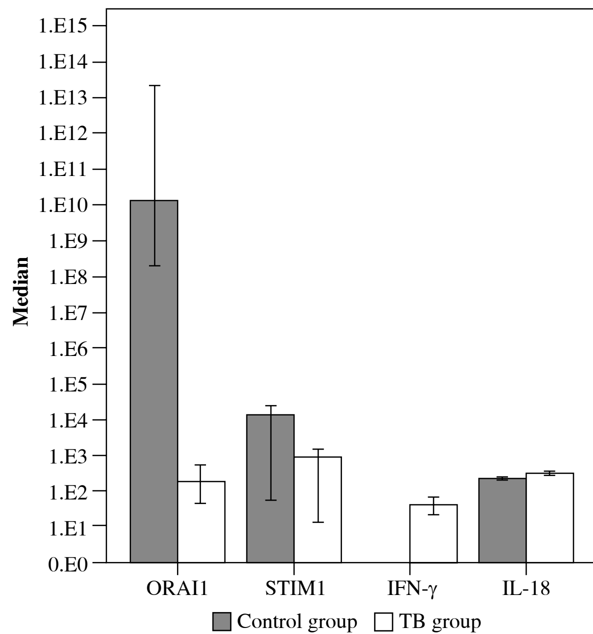

Fig. 1. Differences between groups in mRNA ORAI1, mRNA STIM1, IFN-γ and IL-18 (medians and their 95% CI are presented on a decimal logarithmic scale)

Focusing on *ORAI1* and *STIM1* expression constitutes a limitation of the study because regulators of other Ca^{2+} channels and the intracellular level of Ca^{2+} were not included in the investigation. We assessed the expression of *ORAI1* and *STIM1* genes but did not directly address

Table 2. Spearman correlations among markers in control and tuberculosis (TB) group

Markers	Control group			TB group		
	ORAI1	STIM1	IFN-γ	ORAI1	STIM1	IFN-γ
STIM1	0.07[a]	–		0.12[c]	–	
IFN-γ	–0.18[b]	–0.38[b]	–	–0.10[d]	0.16[d]	–
IL-18	–0.05[b]	0.06[b]	0.13[b]	0.15[e]	–0.03[e]	–0.06[d]

[a] presented for a subsample n = 35, [b] presented for a subsample n = 15, [c] presented for a subsample n = 45, [d] presented for a subsample n = 34, [e] presented for a subsample n = 44

the level of expression of ORAI1 and STIM1 proteins. Moreover, assessing the expression of *ORAI1* and *STIM1* genes and proteins in blood should be performed in a larger group of patients with different severity of TB and should include monitoring of TB dynamics. Further studies should also include a broader range of Ca^{2+} channels and their regulators to investigate their complex functioning in TB patients.

Conclusions

It can be concluded that patients with TB have a lower level of *ORAI1* expression in blood than individuals without TB. It indicates some impairment in the SOCE mechanism of immune cells, which is associated with TB. Despite the lack of significant correlations among *ORAI1* and *STIM1* gene expression levels and levels of two inflammatory cytokines, we have demonstrated that a low level of the *ORAI1* gene and high levels of markers of inflammation are presented simultaneously in blood of TB patients.

Acknowledgements

This study was supported by Rīga Stradiņš University, Latvia, Grant No. 21-1/1/2019-4.

The authors declare no conflict of interest.

References

1. Feske S (2009): ORAI1 and STIM1 deficiency in human and mice: roles of store-operated Ca^{2+} entry in the immune system and beyond. Immunol Rev 231: 189-209.
2. Dheda K, Schwander SK, Zhu B, et al. (2010): The immunology of tuberculosis: from bench to bedside. Respirology 15: 433-450.
3. Desvignes L, Weidinger C, Shaw P, et al. (2015): STIM1 controls T cell–mediated immune regulation and inflammation in chronic infection. J Clin Invest 125: 2347-2362.
4. Demaurex N, Nunes P (2016). The role of STIM and ORAI proteins in phagocytic immune cells. Am J Physiol Cell Physiol 310: C496-C508.
5. Jayachandran R, Sundaramurthy V, Combaluzier B, et al. (2007): Survival of mycobacteria in macrophages is mediated by coronin 1-dependent activation of calcineurin. Cell 130: 37-50.
6. Gupta S, Salam N, Srivastava V, et al. (2009): Voltage gated calcium channels negatively regulate protective immunity to Mycobacterium tuberculosis. PLoS One 4: e5305.
7. Gupta D, Sharma S, Singhal J, et al. (2010): Suppression of TLR2-induced IL-12, reactive oxygen species, and inducible nitric oxide synthase expression by Mycobacterium tuberculosis antigens expressed inside macrophages during the course of infection. J Immunol 184: 5444-5455.
8. Sharma D, Tiwari BK, Mehto S, et al. (2016): Suppression of protective responses upon activation of L-type voltage gated calcium channel in macrophages during Mycobacterium bovis BCG infection. PLoS One 11: e0163845.
9. Sharma S, Meena LS (2017): Potential of Ca^{2+} in Mycobacterium tuberculosis H37Rv pathogenesis and survival. Appl Biochem Biotechnol 181: 762-771.
10. Feske S, Wulff H, Skolnik EY (2015): Ion channels in innate and adaptive immunity. Annu Rev Immunol 33: 291-353.
11. Samarani S, Allam O, Sagala P, et al. (2016): Imbalanced production of IL-18 and its antagonist in human diseases, and its implications for HIV-1 infection. Cytokine 82: 38-51.
12. Keyel PA (2014): How is inflammation initiated? Individual influences of IL-1, IL-18 and HMGB1. Cytokine 69: 136-145.
13. Tan HY, Yong YK, Andrade BB, et al. (2015): Plasma interleukin-18 levels are a biomarker of innate immune responses that predict and characterize tuberculosis-associated immune reconstitution inflammatory syndrome. AIDS 29: 421-431.
14. Feske S, Picard C, Fischer A (2010): Immunodeficiency due to mutations in ORAI1 and STIM1. Clin Immunol 35: 169-182.
15. Zhang X, Guo J, Fan S, et al. (2013): Screening and identification of six serum microRNAs as novel potential combination biomarkers for pulmonary tuberculosis diagnosis. PLoS One 8: e81076.
16. Kumar R, Sahu SK, Kumar M, et al. (2016): MicroRNA17-5p regulates autophagy in Mycobacterium tuberculosis-infected macrophages by targeting Mcl-1 and STAT3. Cell Microbiol 18: 679-691.
17. Ouimet M, Koster S, Sakowski E, et al. (2016): Mycobacterium tuberculosis induces the miR-33 locus to reprogram autophagy and host lipid metabolism. Nat Immunol 17: 677-686.
18. Liu F, Chen J, Wang P, et al. (2018): MicroRNA-27a controls the intracellular survival of Mycobacterium tuberculosis by regulating calcium-associated autophagy. Nat Commun 9: 4295.
19. Chang WC, Lee CH, Hirota T, et al. (2012): ORAI1 genetic polymorphisms associated with the susceptibility of atopic dermatitis in Japanese and Taiwanese populations. PLoS One 7: e29387.
20. McCarl CA, Picard C, Khalil S, et al. (2009): ORAI1 deficiency and lack of store-operated Ca^{2+} entry cause immunodeficiency, myopathy and ectodermal dysplasia. J Allergy Clin Immunol 124: 1311-1318.

INDEX OF PAPERS

Volume 46, Issue 1-2, 2021

Billert H, Bednarek E, Kusza K, Ponichter M, Kurpisz M: Effect of acute isooxic hypercapnia on oxidative activity of systemic neutrophils in endotoxemic rabbits 47

Bogucka-Fedorczuk A, Czyż A, Szuba A, Machnicki MM, Pępek M, Płoski R, Stokłosa T, Wróbel T: Co-occurrence of unclassified myeloproliferative neoplasm and giant cell arteritis in a patient treated with allogeneic hematopoietic stem cell transplantation: a case report and literature review 121

Ding J, Zhang X, Xue J, Fang L, Ban C, Song B, Wu L: CircNPM1 strengthens Adriamycin resistance in acute myeloid leukemia by mediating the miR-345-5p/FZD5 pathway 162

Džopalić T, Božić-Nedeljković B, Jurišić V: The role of vitamin A and vitamin D in modulation of the immune response with a focus on innate lymphoid cells 264

Fasshauer M, Schuermann G, Gebert N, von Bernuth H, Goldacker S, Krueger R, Manzey P, Notheis G, Ritterbusch H, Schauer U, Schulze I, Umlauf V, Widmann S, Baumann U: A structured patient empowerment programme for primary immunodeficiency significantly improves general and health-related quality of life 244

Ferreira S, Masi J, Giménez V, Carpinelli MM, Laterza O, Hermoso M, Ortiz-Villalba J, Chamorro ME, Langjahr P: Effect of gluten-free diet on levels of soluble CD14 and lipopolysaccharide-binding protein in adult patients with celiac disease 225

Glushkov A, Polenok E, Gordeeva L, Mun S, Kostyanko M, Antonov A, Verzhbitskaya N, Vafin I: Immuno-hormonal network in postmenopausal women: disturbance in breast cancer patients 68

Gowin E, Bąbol-Pokora K, Januszkiewicz-Lewandowska D: Mutation in the proline-serine-threonine phosphatase-interacting protein 1 (*PSTPIP1*) gene in a patient with acute lymphoblastic leukemia 270

Hyla-Klekot L, Wolny A, Janas-Kozik M, Koszutski T: Anorexia nervosa and juvenile lupus erythematosus in a 16-year-old female patient – common disease origin or random coincidence? 127

Jałowska MD, Gornowicz-Porowska J, Seraszek-Jaros A, Bowszyc-Dmochowska M, Kaczmarek E, Dmochowski M: Conceptualization and validation of an innovative direct immunofluorescence technique utilizing fluorescein conjugate against IgG + IgG4 for routinely diagnosing autoimmune bullous dermatoses 183

Kaźmierczyk-Winciorek M, Nędzi-Góra M, Słotwińska SM: The immunomodulating role of probiotics in the prevention and treatment of oral diseases 99

Kolesova O, Kramica K, Kolesovs A, Eglite J: Expression of *ORAI1* and *STIM1* genes in blood of patients with pulmonary tuberculosis 275

Kołtan S, Kołtan A, Soszyńska K, Matiakowska K, Morgut-Klimkowska M, Grześk E, Grześk G, Dąbrowska A, Urbańczyk A, Konieczek J, Styczyński J, Haus O, Wysocki M: Killer-cell immunoglobulin-like receptor genotype and haplotype combinations in children treated for acute lymphoblastic leukemia 210

Kostic M, Dzopalic T, Marjanovic G, Urosevic I, Milosevic I: Immunomodulatory effects of galectin-1 in patients with chronic lymphocytic leukemia 54

Kuźmicka W, Moskalik A, Manda-Handzlik A, Demkow U, Wachowska M, Ciepiela O: Influence of iron- and zinc-chelating agents on neutrophil extracellular trap formation 135

Lu Y, Wang G, Li C: Expression of peripheral monocytic programmed death ligand-1 in severe sepsis combined with HBV-related cirrhosis. A pilot observational study 217

Marcinkiewicz J, Witkowski JM, Olszanecki R: The dual role of the immune system in the course of COVID-19. The fatal impact of the aging immune system 1

Mizerska-Wasiak M, Gajewski Ł, Cichoń-Kawa K, Siejko A, Małdyk J, Spława-Neyman A, Zachwieja J, Firszt-Adamczyk A, Stankiewicz R, Drożyńska-Duklas M, Żurowska A, Bieniaś B, Sikora P, Pukajło-Marczyk A, Zwolińska D, Szczepańska M, Pawlak-Bratkowska M, Tkaczyk M, Stelmaszczyk-Emmel A, Pańczyk-Tomaszewska M: Relationship between Gd-IgA1 and TNFR1 in IgA nephropathy and IgA vasculitis nephritis in children – multicenter study 199

Nędzi-Góra M, Górska R, Górski B: The utility of gingival crevicular fluid matrix metalloproteinase-8 provides site-specific diagnostic value for periodontal grading 236

Pawlik-Gwozdecka D, Górska-Ponikowska M, Adamkiewicz-Drożyńska E, Niedźwiecki M: Serum heat shock protein 90 as a future predictive biomarker in childhood acute lymphoblastic leukemia 63

Pérez-Soto E, Oros-Pantoja R, Fernández-Martínez E, Carbonell-Campos JM, Sánchez Monroy V: Seminal pro-inflammatory cytokines and pH are affected by *Chlamydia* infection in asymptomatic patients with teratozoospermia 76

Samelska K, Zaleska-Żmijewska A, Bałan B, Grąbczewski A, Szaflik JP, Kubiak AJ, Skopiński P: Immunological and molecular basics of the primary open angle glaucoma pathomechanism 111

Shoji S, Uchida K, Inoue G, Takata K, Mukai M, Aikawa J, Iwase D, Takano S, Sekiguchi H, Takaso M: Increase in CD5L expression in the synovial membrane of knee osteoarthritis patients with obesity 231

Skopiński P, Radomska-Leśniewska DM, Izdebska J, Kamińska A, Kupis M, Kubiak AJ, Samelska K: New perspectives of immunomodulation and neuroprotection in glaucoma 105

Słotwiński R, Lech G, Słotwińska SM: Molecular aspects of pancreatic cancer: focus on reprogrammed metabolism in a nutrient-deficient environment and potential therapeutic targets 258

Stelmasiak M, Bałan BJ, Mikaszewska-Sokolewicz M, Niewiński G, Kosałka K, Szczepanowska E, Słotwiński R: The relationship between the degree of malnutrition and changes in selected parameters of the immune response in critically ill patients 82

Sun M, Wu J, Liu W: Profiling changes in microRNAs of immature dendritic cells differentiated from human monocytes 10

Švajger U, Rožman PJ: Mixed cultures of allogeneic dendritic cells are phenotypically and functionally stable – a potential for primary cell-based “off the shelf” product generation 152

Volokha A, Bondarenko A, Chernyshova L, Hilfanova A, Stepanovskiy Y, Boyarchuk O, Kostyuchenko L: Impact of the J Project on progress of primary immunodeficiency care in Ukraine 250

Wielińska J, Tarassi K, Iwaszko M, Kościńska K, Wysoczańska B, Mole E, Kitsiou V, Świerkot J, Kolossa K, Kouniaki D, Athanassiades T, Tsirogianni A, Bogunia-Kubik K: Shared epitope and polymorphism of *MICA* and *NKG2D* encoding genes in Greek and Polish patients with rheumatoid arthritis 92

Xu W, Li S, Chang X: E2F2 stimulates CCR4 expression and activates synovial fibroblast-like cells in rheumatoid arthritis 27

Xue X, Liu Q, Xu W, Yuan J, Zhou H, Zou X, Han S, Meng X, Wang X: Imbalanced Th17/Treg in peripheral blood of adult patients with immunoglobulin A vasculitis nephritis 191

Zdanowicz K, Daniluk U, Jewsiejenko E, Krasnodębska M, Motkowski R, Lebensztejn DM: Diagnosis of autoimmune neutropenia in a 10-month-old boy – a case report 118

Zhang B, Zhang Y, Li R, Li Y, Yan W: Knockdown of circular RNA hsa_circ_0003204 inhibits oxidative stress and apoptosis through the miR-330-5p/Nod2 axis to ameliorate endothelial cell injury induced by low-density lipoprotein 140

Zhang X, Zhang J, Li F, Luo Y, Jiang S: PDCD4-mediated downregulation of *Listeria monocytogenes* burden in macrophages 38

Zhang Y, Xie L, Lu W, Lv J, Li Y, Shao Y, Sun J: LncRNA MIAT enhances systemic lupus erythematosus by upregulating CFHR5 expression via miR-222 degradation 17

INDEX OF AUTHORS

Volume 46, Issue 1-2, 2021

Adamkiewicz-Drożyńska E 63
Aikawa J 231
Antonov A 68
Athanassiades T 92
Bałan BJ 82, 111
Ban C 162
Baumann U 244
Bąbol-Pokora K 270
Bednarek E 47
Bieniaś B 199
Billert H 47
Bogucka-Fedorczuk A 121
Bogunia-Kubik 92
Bondarenko A 250
Bowszyc-Dmochowska M 183
Boyarchuk O 250
Božić-Nedeljković B 264
Carbonell-Campos JM 76
Carpinelli MM 225
Chamorro ME 225
Chang X 27
Chernyshova L 250
Cichoń-Kawa K 199
Ciepiela O 135
Czyż A 121
Daniluk U 118
Dąbrowska A 210
Demkow U 135
Ding J 162
Dmochowski M 183
Drożyńska-Duklas M 199
Džopalić T 54, 264
Eglite J 275
Fang L 162
Fasshauer M 244
Fernández-Martínez E 76
Ferreira S 225
Firszt-Adamczyk A 199
Gajewski Ł 199
Gebert N 244
Giménez V 225
Glushkov A 68
Goldacker S 244
Gordeeva L 68
Gornowicz-Porowska J 183
Gowin E 270
Górska R 236
Górska-Ponikowska M 63
Górski B 236
Grąbczewski A 111
Grześk E 210
Grześk G 210
Han S 191
Haus O 210
Hermoso M 225
Hilfanova A 250
Hyla-Klekot L 127
Inoue G 231
Iwase D 231
Iwaszko M 92
Izdebska J 105
Jałowska MD 183
Janas-Kozik M 127
Januszkiewicz-Lewandowska D 270
Jewsiejenko E 118
Jiang S 38
Jurišić V 264
Kaczmarek E 183
Kamińska A 105
Kaźmierczyk-Winciorek M 99
Kitsiou V 92
Kolesova O 275
Kolesovs A 275
Kolossa K 92
Kołtan A 210
Kołtan S 210
Konieczek J 210
Kosałka K 82
Kostic M 54
Kostyanko M 68
Kostyuchenko L 250
Koszutski T 127
Kościńska K 92
Kouniaki D 92
Kramica K 275
Krasnodębska M 118
Krueger R 244
Kubiak AJ 105, 111
Kupis M 105
Kurpisz M 47
Kusza K 47
Kuźmicka W 135
Langjahr P 225
Laterza O 225
Lebensztejn DM 118
Lech G 258
Li C 217
Li F 38
Li R 140
Li S 27
Li Y 140
Li Y 17
Liu Q 191
Liu W 10
Lu W 17
Lu Y 217
Luo Y 38
Lv J 17
Machnicki MM 121
Małdyk J 199
Manda-Handzlik A 135
Manzey P 244
Marcinkiewicz J 1

Marjanovic G 54
Masi J 225
Matiakowska K 210
Meng X 191
Mikaszewska-Sokolewicz M 82
Milosevic I 54
Mizerska-Wasiak M 199
Mole E 92
Morgut-Klimkowska M 210
Moskalik A 135
Motkowski 118
Mukai M 231
Mun S 68
Nędzi-Góra M 99, 236
Niedźwiecki M 63
Niewiński G 82
Notheis G 244
Olszanecki R 1
Oros-Pantoja R 76
Ortiz-Villalba J 225
Pańczyk-Tomaszewska M 199
Pawlak-Bratkowska M 199
Pawlik-Gwozdecka D 63
Pérez-Soto E 76
Pępek M 121
Płoski R 121
Polenok E 68
Ponichter M 47
Pukajło-Marczyk A 199
Radomska-Leśniewska DM 105
Ritterbusch H 244
Rožman PJ 152
Samelska K 105, 111
Sánchez Monroy V 76
Schauer U 244
Schuermann G 244
Schulze I 244
Sekiguchi H 231
Seraszek-Jaros A 183
Shao Y 17
Shoji S 231
Siejko A 199
Sikora P 199
Skopiński P 105, 111
Słotwińska SM 99, 258
Słotwiński R 82, 258
Song B 162
Soszyńska K 210
Spława-Neyman A 199
Stankiewicz R 199
Stelmasiak M 82
Stelmaszczyk-Emmel A 199
Stepanovskiy Y 250
Stokłosa T 121
Styczyński J 210
Sun J 17
Sun M 10
Švajger U 152
Szaflik JP 111
Szczepanowska E 82
Szczepańska M 199
Szuba A 121
Świerkot J 92
Takano S 231
Takaso M 231
Takata K 231
Tarassi K 92
Tkaczyk M 199
Tsirogianni A 92
Uchida K 231
Umlauf V 244
Urbańczyk A 210
Urosevic I 54
Vafin I 68
Verzhbitskaya N 68
Volokha A 250
von Bernuth H 244
Wachowska M 135
Wang G 217
Wang X 191
Widmann S 244
Wielińska J 92
Witkowski JM 1
Wolny A 127
Wróbel T 121
Wu J 10
Wu L 162
Wysocki M 210
Wysoczańska B 92
Xie L 17
Xu W[encheng] 191
Xu W[anju] 27
Xue J 162
Xue X 191
Yan W 140
Yuan J 191
Zachwieja J 199
Zaleska-Żmijewska A 111
Zdanowicz K 118
Zhang B 140
Zhang J 38
Zhang X[iaochun] 162
Zhang X[ingju] 38
Zhang Y 140
Zhang Y 17
Zhou H 191
Zou X 191
Zwolińska D 199
Żurowska A 199