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RESEARCH REPORT 2016

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DEPARTMENT OF EXPERIMENTAL ONCOLOGY
Head: Professor Leon Strządała, Ph.D.

Laboratory of Experimental Anticancer Therapy
Head: Professor Joanna Wietrzyk, Ph.D.

Studies on the mechanisms of tumor progression and metastasis and on the effects of experimental antitumor therapy

Regulation of the expression of VDR receptor in human breast cancer and normal breast epithelial cells by miR-125b

The aim of the experiments was to examine whether miR-125b regulates the expression of the vitamin D receptor (VDR) in cell lines of normal and malignant epithelium derived from human mammary gland.

Previous studies have shown that MCF-7 cells are sensitive to the action of the calcitriol analog PRI-2191 in contrast to MDA-MB-231 and MCF-10A. Analyses of expression of VDR and $1,25D_3$-MARRS showed that all cell lines tested express VDR and MARRS at the mRNA level. In MCF-7 we observed the highest levels of VDR protein from lines tested, although the highest level of mRNA was not seen. The highest levels of VDR mRNA were observed in MCF-10A. The analysis of the expression of miR-125b showed that all cell lines tested expressed miR-125b, the highest level being observed in MDA-MB-231 cells, and the lowest in the most sensitive cell line, MCF-7. An inverse correlation was observed according to VDR protein level, i.e. low miR125b correlated with high levels of the VDR protein.

Studies on the potential application of bisphosphonates in anti-cancer treatment

In 2016, studies on the anti-proliferative activity of two new bisphosphonates, WG12399C and WG12592A, against cancer cells originating from organs other than breast were conducted. The bisphosphonate 12592A exerted significant and specific anti-proliferative activity against SKOV-3 and Hs294T cells. As we previously demonstrated, the new bisphosphonates showed potent anti-proliferative activity towards murine macrophages J774E and RAW264.7. The conditions for effective differentiation of macrophages into osteoclasts were established and the studies on the potential anti-osteoporotic properties of the new bisphosphonates were started. Our results show that the potential clinical application of bisphosphonates may not be limited to breast cancer and metastases to bones. They also indicate that the two new bisphosphonates possess different molecular mechanisms of action, even though both compounds belong to the same group of N-bisphosphonates.

Evaluation of the effect of vaccines based on cells transduced with IL-2 and IL-12 lentiviral carriers on the tumor environment in mouse colon carcinoma models (continuation)

The purpose of our task was to elaborate strategies, using application of anti-cancer vaccines based on genetically modified cells, supporting the renewal and/or modulation of the immune response after the cytoreductive impact of conventional therapy. We used third generation lentiviral vectors encoding sequences of IL-2 and IL-12 genes as well as the IL-15 gene for modification of murine tumor cell lines and dendritic cells of the JAWS II line. The introduction of a vector containing genes encoding IL-2 or IL-15 had no effect on the ability of transductants for the production of other cytokines, excluding transductants of TC-1 cells, whose production of IL-6 decreased compared to wild type cells. The effect of transduction on changes of MHC class I expression was observed. The use of tumor antigens and supernatants from above transduced tumor TC-1 or X63-Ag 8.653 cell culture (TAgSup), for *in vitro* stimulation, induced an increase in CD11c, CD40, CD80, and CD86 expression on the surface of dendritic cells, while the level of MHC class II antigen expression depended on the type of stimulators. Pre-activated BM-DC cells used in the four-day mixed culture with spleen cells caused a change in the size of their subpopulations. Culture of splenocytes with BM-DC stimulated with TC-1 cell TAgSup resulted in an increase in the percentage of CD8 +, CD4 +, and CD19 + and a decrease in CD49b + splenocyte percentage. In contrast, BM-DCs with X63-Ag8.653 cell TAgSup induced an increase of the percentage of CD19+ cells and reduced the size of CD8+, CD4+, and CD49b+ subpopulations. The most important achievement of the study is the implementation of new techniques, extending the scope of the research in order to enrich the study in thematic projects.

Laboratory of Tumor Molecular Immunobiology
Head: Professor Leon Strządała, Ph.D.

Molecular mechanism of programmed cell death and proliferation in normal and tumor cells

Currently, 5-fluorouracil, irinotecan (also known as CPT-11), and oxaliplatin constitute the backbone of chemotherapy for colorectal cancer (CRC). Unfortunately, the molecular heterogeneity of CRC creates considerable variability in response to treatment among patients with the same disease stage. Because the currently approved therapies have failed in a substantial number of CRC patients, new efficient drug combinations are constantly being sought. Emerging data indicate that 5-azanucleosides are able to sensitize cancer cells to the standard chemotherapeutic agents and contribute to overcoming intrinsic or acquired chemoresistance.

It is now well established that genetic mutations cooperate with epigenetic changes to drive the formation and progression of normal colorectal epithelium into adenocarcinomas. Abnormalities in DNA methylation occur early in cancer progression, even before the appearance of the aberrant crypt foci (ACF), the first neoplastic lesions identified in CRC formation.

The reversibility of epigenetic modifications makes them attractive targets for cancer treatment. The two most extensively studied inhibitors of DNA methylation are 5-azacytidine (5-aza-C) and 5-aza-2'-deoxycytidine (5-aza-dC, decitabine), analogs of cytidine and 2'-deoxycytidine, respectively, with a nitrogen atom replacing the carbon at the 5 position of the cytosine base.
CRC cells with different genetic backgrounds (HCT116, DLD-1, HT-29) were sequentially treated with 5-azanucleosides and topoisomerase inhibitors. The combined effects of these two drug classes on cell viability, apoptosis, signaling pathways, and colony formation were investigated.

We have demonstrated that pretreatment with DNA demethylating agents, 5-aza-2'-deoxycytidine and 5-azacytidine, sensitizes CRC cells to topoisomerase inhibitors (irinotecan, etoposide, doxorubicin, mitoxantrone), reducing cell viability and clonogenicity and increasing apoptosis more effectively than individual compounds at the same or even higher concentrations. 5-Azanucleosides did not cause considerable immediate toxic effects as evaluated by analysis of cell viability, apoptosis, DNA damage (γH2A.X), and endoplasmic reticulum (ER) stress (CHOP). However, 5-azanucleosides exerted long-lasting effects, reducing cell viability, changing cell morphology, and affecting the phosphoinositide 3-kinase (PI3-kinase)/Akt signaling pathway. We found that single exposure to 5-azanucleosides is sufficient to induce long-lasting sensitization to topoisomerase inhibitors. The combinatorial, but not separate, treatment with low doses of 5-aza-2'-deoxycytidine (0.1 μM) and etoposide (0.5 μM) caused a long-lasting (almost 70 days) reduction in clonogenic/replating ability of DLD-1 cells.

Our studies have shown that pretreatment of CRC cells with 5-azanucleosides potentiates the anticancer effects of topoisomerase inhibitors, suggesting that the combination of these two drug classes represents a promising therapeutic approach for the treatment of CRC and possibly other cancers. Importantly, prior exposure to 5-azanucleosides could potentially reduce topoisomerase inhibitors dosing and therefore decrease their side effects, such as myelosuppression. Similarly, it has been reported that 5-aza-C potentiates anticancer activity of cisplatin and, at the same time, attenuates the cisplatin-induced nephrotoxicity. Thus, our findings strongly encourage future *in vivo* studies on combinatorial use of DNA demethylating agents and topoisomerase inhibitors.

Laboratory of Biomedical Chemistry
Head: Professor Janusz Boratyński, Ph.D., Eng.

Protein-boron conjugates

1. Boron clusters represent a vast family of boron-rich compounds with extraordinary properties that provide the opportunity of exploitation in different areas of chemistry and biology. In addition, boron clusters are clinically used in the boron neutron capture therapy (BNCT) of tumors. In this

paper, a novel, solid-state (solvent-free), thermal method for protein modification with boron clusters is proposed. The method is based on a cyclic ether ring opening in an oxonium adduct of cyclic ether and a boron cluster with nucleophilic centers of the protein. Lysozyme was used as the model protein, and the physicochemical and biological properties of the obtained conjugates were characterized.

The main residues of modification were identified as arginine-128 and threonine-51. No significant changes in the secondary or tertiary structures of the protein after tethering of the boron cluster were found by mass spectrometry and circular dichroism measurements. However, some changes in the intermolecular interactions and hydrodynamic and catalytic properties were observed.

To the best of our knowledge, we have described the first example of an application of cyclic ether ring opening in the oxonium adducts of a boron cluster for protein modification. In addition, a distinctive feature of the proposed approach is performing the reaction in a solid state and at an elevated temperature.

The proposed methodology provides a new route to protein modification with boron clusters and extends the range of innovative molecules available for biological and medical testing.

2. Two complementary methods, “in solution” and “in solid state”, for the synthesis of lysozyme modified with metallacarborane (cobalt bis(dicarbollide), Co(C2 B9 H11)2 (2-)) were developed. As metallacarborane donors, oxonium adducts of cobalt bis(dicarbollide) and 1,4-dioxane or tetrahydropyran were used. The physicochemical and biochemical properties of the obtained lysozyme-metallacarborane conjugates were studied for changes in secondary and tertiary structure, aggregation behavior, and biological activity. Only minor changes in primary, secondary, and tertiary protein structure were observed, caused by the single substitution of metallacarborane on lysozyme. However, the modification produced significant changes in lysozyme enzymatic activity and a tendency toward time- and temperature-dependent aggregation.

Removal of endotoxins from bacteriophage preparations

The method of removing pyrogen contaminating bacteriophage preparations by water immiscible solvent extraction was developed. During this process most of the phage lytic activity is retained in the aqueous phase, while endotoxin accumulates in the organic solvent. The levels of endotoxin in the aqueous bacteriophage-containing fraction determined by limulus amebocyte lysate or EndoLISA assay were exceptionally low. While the initial endotoxin levels in the crude phage lysates ranged between 10^3 and 10^5 EU/ml, the average level after organic extraction remaining in the aqueous fraction was 5.3 EU/ml. The purification procedure is scalable, efficient and applicable to all the bacteriophages tested: T4, HAP1 (*E. coli*) and F8 (*P. aeruginosa*).

DEPARTMENT OF CLINICAL IMMUNOLOGY

Laboratory of Clinical Immunogenetics and Pharmacogenetics
Head: Professor Katarzyna Bogunia-Kubik, Ph.D.

Polymorphism and expression of pro-inflammatory factors in patients with rheumatic diseases treated with TNF-alpha inhibitors

MicroRNA-146a (miR-146a) plays an important role in the regulation of inflammatory innate immune responses, and can be differentially expressed in rheumatoid arthritis (RA). Through the NF-κB pathway, this molecule is able to stimulate the release of pro-inflammatory cytokines such as TNF-α, IL-1β, and IL-17. Our previous studies showed that genetic variability of genes coding for IL-17A, IL-17F, and TNF-α and its receptor can affect RA susceptibility and response to TNF-α inhibitors (TNFi) (*Arch Immunol Ther Exp.* 2015, *Joint Bone Spine* 2015). It was also documented that single-nucleotide polymorphisms (SNPs) in miRNA sequences may alter miRNA expression and that miR-146a rs2910164 SNP may contribute to RA development. Thus, in addition to SNPs within IL-66 and its receptor encoding gene (*IL6*: rs1800795, C>G; *IL6R*: rs2228145, A>C, Asp358Ala), the miR-146a-3p (rs2910164, G>C) and *NFkB1* (rs28362491, ins/del ATTG) polymorphisms were studied. Moreover, miR-146a-5p expression in patients' sera as well as IL-6 and CRP serum levels was assessed. Detected genotypes and expression levels were analyzed to assess their relationship with clinical outcome of treatment as well as predisposition to the disease. None of the polymorphisms studied was found to be associated with disease susceptibility.

RA patients carrying the *NFkB1 ins/ins* genotype were characterized by a worse response to TNFi treatment (p=0.023). The miR-146a-5p expression levels were lower in patients before TNFi therapy as compared to those measured after three months of treatment (p=0.033) and those observed in healthy controls (p=0.048). Moreover, patients with higher circulating miR-146a-5p levels after three months of TNFi administration were more frequently carrying the rs2910164-C allele (p=0.032). These results support the hypothesis that miR-146a might be involved in pathogenesis of RA and imply that miR-146a-3p polymorphism may be associated with miR-146a-5p levels in serum after anti-TNF-α treatment. We did not observe any significant relationship between the *NFkB1* polymorphism and miR-146a-5p expression (*Arch Immunol Ther Exp. in press*).

In psoriatic arthritis (PsA) patients an association between the *IL6* SNP and efficacy of methotrexate (MTX) treatment was seen. The *IL6 G* carriers were worse responders to MTX therapy than *CC* homozygotes (p=0.046). Moreover, *IL6* polymorphism correlated with IL-6 and CRP serum levels. Patients with *GG* homozygous genotype exhibited higher IL-6 serum levels

(p=0.032), while IL-6 concentrations were significantly higher in *G* positive patients than *CC* homozygotes (p=0.026). No significant relationships were observed with the *IL6R* SNP rs2228145.

Cereblon-coding gene polymorphism and its role in lenalidomide therapy in patients with multiple myeloma

Lenalidomide (Revlimid; a derivative of thalidomide) is one of the immunomodulatory drugs (IMIDs) used in multiple myeloma treatment. It is, however, ineffective in some patients. Recent research suggests that cereblon (CRBN) has a major role in metabolism of immunomodulatory drugs and that *CRBN* gene polymorphism influences lenalidomide treatment effectiveness. The research objective is to determine polymorphic variants (that have not been studied in the context of any hematological or neoplastic diseases) of the gene coding for CRBN and to analyze their association with response to lenalidomide treatment in multiple myeloma patients. Genotyping for *CRBN* rs711613 A>G and rs1045433 A>G was performed using the LightSNiP assay and melting curve analysis.

Our previous studies (*Leuk Res* 2015) did not show any relation between *CRBN* rs121918368 C>T and either disease, or a response to treatment (we did not detect polymorphic variants of this gene in our population). Subsequently, analyzing two other polymorphic sites in the *CRBN* gene (*Arch Immunol Ther Exp. in press*), we found the following: (i) *CRBN* rs711613 *A* allele was associated with remission after first line therapy (p=0.010), this association being independent of age, gender and stage of disease; (ii) additionally, the *CRBN* rs711613 *A* allele was associated with remission in a subset of patients treated with thalidomide (p=0.023); (iii) the *CRBN* rs1045433 *G* variant was more common in patients with remission after first line therapy (p=0.081); (iv) the rs711613 *G* allele was less common in patients in the advanced stage III of disease (Durie-Salmon classification system; p=0.005).
These results might suggest that *CRBN* polymorphisms can affect the outcome of IMID treatment in multiple myeloma patients.

Laboratory of Immunogenetics and Tissue Immunology
Head: Professor Piotr Kuśnierczyk, Ph.D.

Single nucleotide polymorphisms of the ERAP1 gene and risk of NSCLC: a comparison of genetically distant populations, Chinese and Caucasian

An effective cytotoxic immune response to neoplastic cells requires efficient presentation of antigenic peptides to T lymphocytes by HLA class I (HLA-I) molecules. The HLA-I-bound peptide repertoire depends on antigen-processing machinery molecules. Aminopeptidase residing in endoplasmic reticulum 1 (ERAP1) trims peptides to the optimal length for HLA-I binding. Single

nucleotide polymorphisms (SNPs) in the ERAP1 gene result in changes in aminopeptidase activity and specificity. This may affect susceptibility to cancer. However, non-small cell lung carcinoma (NSCLC) has not been studied in this respect. We compared genotype and haplotype frequencies of four coding, nonsynonymous ERAP1 SNPs, rs26653G>C, rs26618T>C, rs30187C>T, and rs27044C>G, in NSCLC occurring in two genetically distant populations, Chinese and Poles. We found associations of all four SNPs with NSCLC in Chinese but not in Poles. The differences in ERAP1-NSCLC associations might be explained by highly significant differences in SNP genotype frequencies between Chinese and Poles (except for rs26618). In accordance with this, the most frequent ERAP1 haplotypes were distributed differently in cases versus controls in Chinese, but not in Poles. Our findings add to the differences between Orientals and Caucasians in genetics of disease susceptibility.

Possible role of HLA-G, LILRB1 and KIR2DL4 gene polymorphisms in spontaneous miscarriage

The KIR2DL4 receptor and its ligand HLA-G are considered important for fetal-maternal immune tolerance and successful pregnancy. The absence of a particular variant of KIR2DL4 might be a bad prognostic factor for pregnancy outcome. However, it could be compensated by the presence of the respective LILRB1 allele. Therefore, we investigated the KIR2DL4, LILRB1 and HLA-G polymorphisms in 277 couples with spontaneous abortion and 219 control couples by HRM, PCR-SSP and RFLP methods. We found a protective effect of women's heterozygosity in HLA-G rs1233334 (-716C>G>T) and LILRB1 rs41308748 (5651G>A) against spontaneous abortion. Surprisingly, we observed more 9A/10A genotypes of KIR2DL4 in the group of male partners from the miscarriage group in comparison to the men from the control group. In contrast, there was no association of women's KIR2DL4 polymorphism with susceptibility to spontaneous abortion. Multivariate analysis confirmed that women's HLA-G -716C>G>T and LILRB1 5651G>A as well as men's KIR2DL4 9A/10A are important in terms of protection against or susceptibility to miscarriage, respectively. In conclusion, a woman's heterozygosity in HLA-G and LILRB1 might be an advantage for success of reproduction, but the partner's heterozygosity in KIR2DL4 might have an opposite effect.

Variants in BAFF and BAFF-R genes and the risk of CLL in the Polish population

The BAFF/BAFF-R axis seems to play an important role in the development and progression of chronic lymphocytic leukemia (CLL). In this case-control study we investigated the association between 8 SNPs in the BAFF and BAFF-R genes with the risk of sporadic CLL in a group of 439 CLL patients and 477 healthy subjects, including assessment of haplotypes and gene×gene interactions. We also examined the correlation between selected SNPs and CLL clinical parameters,

CLL outcome as well as BAFF plasma level and intracellular BAFF expression. Our results point to the possible association of the rs9514828 (– 871C>T) SNP of the BAFF gene (i.e., the – 871 TT genotype) and CLL risk. We also noted the possible association of the rs1041569 (– 2701 A>T) SNP of the BAFF gene (the – 2701 TT genotype) with the risk of CLL. Moreover, we found that subjects with the rs61756766 CT genotype of the BAFF-R gene had two times higher risk of CLL than subjects with CC genotypes. None of the SNPs investigated here were associated with CLL clinical parameters, CLL outcome, BAFF plasma level or intracellular BAFF expression. In conclusion, our results showed that SNPs in BAFF and BAFF-R may be considered as potential risk factors of B-CLL.

Lack of detectable fetal microchimerism in psoriasis vulgaris lesions and in non-affected skin in spite of its presence in peripheral blood CD34-positive and CD34-negative cells

Microchimerism is defined as the stable presence of low numbers of cells derived from a different individual due to cell transfer between twins or between mother and fetus during pregnancy. Fetal cells in the organism of the mother (FMc) are postulated to play a role in autoimmune diseases. Psoriasis is a disease which has an autoimmune component, but no study on microchimerism in this disease has been reported. The easiest way to detect microchimerism is to look for male cells in blood or other tissues of a woman who previously delivered a son. Here, we looked for the presence of male cells in mononuclear cell subpopulations from peripheral blood and in skin samples of women with psoriasis and of healthy women. We detected FMc in similar proportions of patients and controls in CD4+, CD8+ and CD34+ cells, whereas in CD34- cells they were present in a higher fraction of controls, and a similar but non-significant difference was observed in CD19+ cells. No microchimeric cells were detected in patients' skin samples, either from affected or non-affected skin, or in skin tissue from healthy control individuals. Therefore, our result does not prove the involvement of microchimerism in the etiology of psoriasis.

Laboratory of Clinical Immunology
Head: Professor Andrzej Lange, M.D.

The Laboratory of Clinical Immunology focused its research activity on the following projects:

1. Characteristic of mesenchymal stem cells (MSC) expanded from the human bone marrow cell population. The results of the study are illustrated below and proved that MSC cultured in the bioreactor have the potential to differentiate into dedicated lineages. Five cultures were performed with reproducible results.

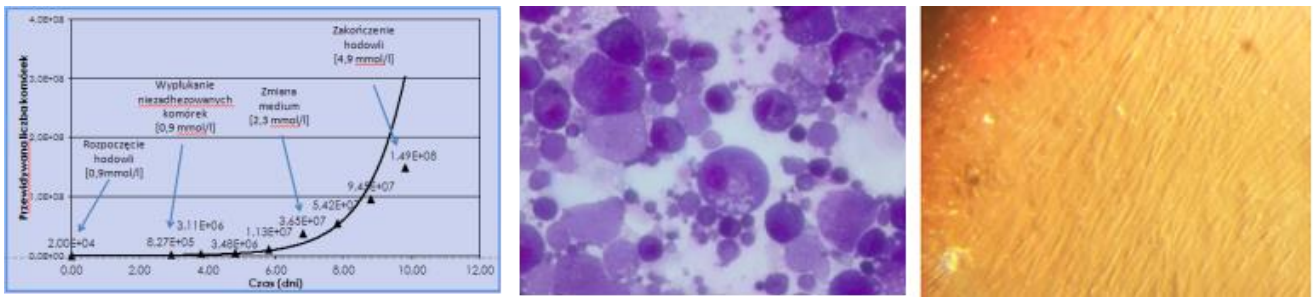

Figure 1. Fresh bone marrow suspension cultured in the bioreactor in a closed system for differentiation to mesenchymal stem cells, from left to right: growth curve of cells expressed as an increase in the medium lactate content, cells harvested after the culture seen in light and inverted microscope.

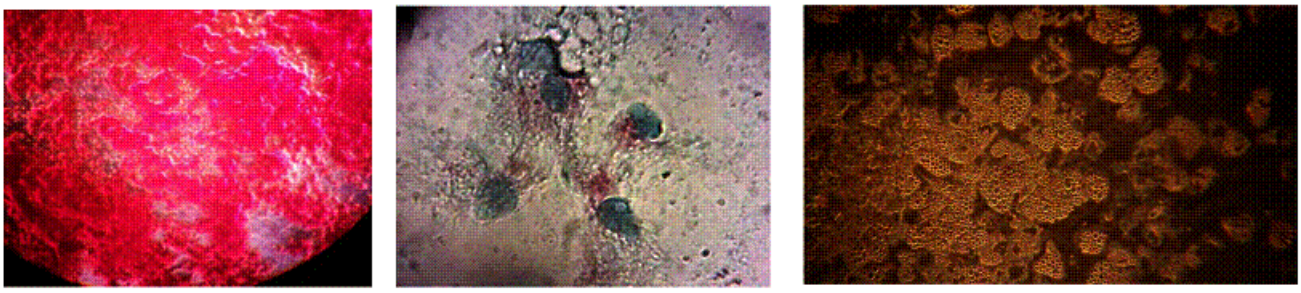

Figure 2. MSC exert suppressive activity and if cultured in appropriate conditions differentiate into osteocytes, chondrocyte and adipocytes (from left to right).

In three experiments, it was found that MSC are able to decrease the response of lymphocytes to T cell activators in 7-day culture.

2. The technique based on centrifugation in a special chamber (commercially available) in autologous plasma was investigated as to the profile of centrifuged cells. It was found that the cell suspensions after centrifugation were enriched in mononuclear cells and of note in those having stemness characteristics (positivity for CD90, CD105, CD73), as well as in the cells having CD133 and being Aldefluor-positive. Altogether 10 experiments were performed, with reproducible results. In addition, microbial quality control was performed in all marrow collections, and proved that the procedure was safe.

3. The cells obtained as above in point 2) were injected into the knees or the hips in 10 patients who suffered from degenerative arthritis. In all cases a clinical improvement was documented, as assessed by standardized questionnaires. Thus the procedure proved to be effective in enrichment of stem cells, showing, in concert with other marrow-derived cells infused, a positive anti-phlogistic and regenerative potential.

4. The new procedure developed within the Innomed/I/1/NCBR/2014 grant was described and successfully used in the clinic. The procedure is based on the use of donor lymphocytes (obtained from primary transplant material or employing leukapheresis). The cells were profiled for the content of lymphocytes and injected using of a trepan directly into the marrow bone spatial areas affected by relapsing leukemia, thus allowing direct contact between donor

lymphocytes and leukemia cells (allorecognition within HLA-matched pair), consequently promoting expansion at the site of relapse. Clinically the procedure, if used for decreasing the blast load, facilitated achieving remission. Of note, the marrow was found to be colonized by CD8+ cells which were CD279 positive. The cells resemble tumor-infiltrating lymphocytes (TILs). The effectiveness of this novel way of delivering donor lymphocytes remains under investigation to document the ability of infused cells to kill leukemia cells.

LABORATORY OF BACTERIOPHAGES
Head: Professor Andrzej Górski, M.D.

Phage neutralization by sera of patients receiving phage therapy

The aim was to study the association between the phage neutralization of patients sera and the clinical outcome of phage therapy (PT). 62 patients with various bacterial infections receiving PT with different routes of phage administration as well as 30 healthy volunteers were studied. The analysis of the association between level of antiphage activity of sera (AAS) during PT and clinical results indicated that the level of AAS is not correlated with the outcome of PT.

In the next study 108 patients with bacterial infections received PT (*S. aureus* phage cocktail or single *S. aureus* phages) with different routes of phage administration. We observed high AAS in sera of 42.9% of patients using locally a cocktail of phages. High AAS was observed in sera of 17.1% of patients who received local monotherapy. Our results suggest that antibody responses to PT may vary depending on whether patients receive monotherapy or a cocktail of phages.

Antibody production in response to staphylococcal MS-1 phage cocktail in patients undergoing phage therapy

In this study, we investigated the humoral immune response (through the release of IgG, IgA, and IgM antiphage antibodies) to a staphylococcal MS-1 phage cocktail in 20 patients undergoing PT orally and/or locally. The majority of patients did not show a noticeably higher level of antiphage antibodies in their sera during phage administration. The presence of increased levels of antiphage antibodies IgG and IgM did not translate into unsatisfactory clinical results of PT. On the other hand, a negative outcome of the treatment occurred in some patients who showed relatively weak production of antiphage antibodies during PT. The outcome of PT does not primarily depend on the appearance of antiphage antibodies in sera of patients during therapy.

Analysis of the results of the application of purified staphylococcal phage cocktail in the treatment of patients with chronic bacterial infections

We retrospectively analyzed the efficacy of the application of a purified staphylococcal phage cocktail (containing P4, A5/80, and 676/Ż phages, each at titer no less than 1×109 pfu/ml) in the treatment of 11 patients with chronic bacterial infections treated at the Phage Therapy Unit of the Medical Centre of the Hirszfeld Institute of Immunology and Experimental Therapy PAS in Wrocław. A good response to the treatment was observed in 54.5% of them, and no adverse events related to this formulation were reported. Although it was not significantly different when compared to the group of 43 patients (good response in 32.6%) who used a non-purified staphylococcal phage cocktail lysate (a mixture of the same phages, each at a titer no less than 5×105 pfu/ml), this trend (40% increase of efficacy) justifies further use of the purified staphylococcal phage cocktail in experimental phage treatment.

T4 phage tail adhesin gp12 counteracts LPS-induced inflammation in vivo

Bacteriophages that attack Gram-negative bacteria commonly bind to the bacterial surface by interaction of specific phage proteins with bacterial lipopolysaccharide. Lipopolysaccharide, also called endotoxin, is a potent inflammation-inducing factor, often responsible for negative and life-threatening physiological effects. Phage short tail fiber proteins (tail adhesin, tailspike, gp12) mediate adsorption of T4-like bacteriophages to *Escherichia coli*, through direct binding of these proteins or LPS. Since LPS is able to exert a major impact on the immune response in animals and in humans, we have tested LPS-binding phage protein gp12 as a potential modulator of the LPS-induced immune response. We have produced tail adhesin gp12 in a bacterial expression system and confirmed its ability to form trimers and to bind lipopolysaccharide *in vitro* by dynamic light scattering. This product had no negative effect on mammalian cell proliferation in vitro. Furthermore, no harmful effects of this protein were observed in mice (100 μg/mouse). Thus, gp12 was used in combination with LPS in a murine model, and it decreased the inflammatory response to LPS in vivo, as assessed by serum levels of cytokines IL-1 alpha and IL-6 and by histopathological analysis of spleen, liver, kidney and lungs. Thus, in future studies gp12 may be considered as a potential tool for modulation and specifically for counteracting LPS-related physiological effects *in vivo*.

LABORATORY OF GLYCOBIOLOGY AND CELLULAR INTERACTIONS
Head: Professor Danuta Duś, Ph.D.

Biology of cells involved in regenerative and neoplastic processes

In 2016 our laboratory continued the investigations on the biology of human endothelial precursor cells (HEPCCB.1 and HEPC-CB.2 cell lines) and mesenchymal/progenitor cells of different tissue origin in the context of regenerative processes. We analyzed the profile and efficacy of cytokines and growth factors secreted by endothelial precursor cells (EPC) in new vessel formation *in vitro* by the mature vessel endothelial cells of the human skin cell line HSkMEC.2. Growth factors released by EPC increased efficacy of new blood vessel formation by the HSkMEC.2 cell line independently of their concentration (50%, 100%) and oxygen level (normoxia, hypoxia). Also co-culture of human endothelial progenitors HEPCCB.1 with the mature endothelial cell line HSkMEC.2 significantly increased the efficacy of blood vessel formation in vitro, compared to monocultures of HEPCCB.1 HSkMEC.2.

Working on the biological properties of mesenchymal stem cells of bone marrow (BM MSC) and adipose tissue (AT MSC) origin, similarly to the previously obtained AT MSC cell line, we created new immortalized BM MSC cell lines using hTERT and pSV402 plasmids. We obtained two different MSC cell lines of bone marrow origin, BM MSC1 and BM MSC2. Both cell lines express basic antigens specific for MSC such as CD73, CD90, and CD105, and both are negative for hematopoietic markers CD45, CD31, and CD34. Both cell lines express HLA class I antigens, but they differ in HLA class II expression: BM MSC1 is HLA-DR negative while BM MSC2 is HLA DR positive. The next step was assessment of biological activity of AT MSC and BM MSC cell lines. Using Microarray Protein Membrane (Custom C-Series Human Cytokine Antibody Array, RayBiotech Inc.) we identified a wide range of cytokines, chemokines and trophic factors produced by both cell lines; however, they differ in the level of produced factors. BM MSC cell lines have greater ability to produce IL-8, VEGF, and IL-13, whereas AT MSC cell lines have greater potential to produce IL-6, osteoprotegerin, and eotaxin.

Looking for alternative sources of MSC, for experimental studies and as potential candidates for tissue regeneration, we analyzed the phenotype and differentiation potential of MSC isolated from human bone marrow (BM), skeletal muscle and skin, collected from limbs amputated due to critical limb ischemia. Adherent cells isolated from BM and those of myogenic and skin origin express phenotypes characteristic for naïve MSC CD73, CD90, and CD105, as confirmed by flow cytometry and immunofluorescence staining, but their expression was downregulated during the follow-up period (after P7). Co-expression of CD73/CD146, CD90/CD146, and CD105/CD146 on the proportion of adherent cells was detected in MSCs originating from BM, skeletal muscle and skin. A fraction of cells expressing CD146 strongly co-expressed PDGFR-α (up to P7). Cells

isolated from all examined tissues were able to differentiate into chondrocytes, osteoblasts and neurons. Moreover, MSC isolated from BM and skin differentiated into adipocytes; however, MSC isolated from skeletal muscle were not capable to form adipocytes. Our observations suggest that MSC isolated from BM and skin biologically represent multipotent cells able to differentiate into different types of tissue, whereas progenitor cells isolated from skeletal muscle have tissue-specific character. The heterogenic nature of MSC isolated from different tissues was confirmed by the presence of subpopulations with phenotypes specific for pro-angiogenic progenitors (expressing CD146, VEGF, and PDGFRα).

Working on tumor cell biology, we are investigating the role of the tumor milieu in anti-tumor therapy. The main goal of the project is to determine the role of IL-10 in activation of the suppressive mechanisms used by tumor-induced myeloid-derived suppressor cells (MDSCs). Research performed in 2016 concerned the construction of lentivectors encoding shRNA sequences specific for IL-10 and IL-10R molecules and estimation of the vectors' efficiency in silencing IL-10 and IL-10R gene expression in MDSCs differentiated in vitro from murine bone marrow. The next stage of our investigations involved preparation of lentivectors suitable for in vivo function which will be applied as one of the components of anti-tumor therapy in MC38 colon carcinoma bearing mice.

Podoplanin increases the migration of human fibroblasts and affects the angiogenesis of endothelial cells. The possible role of cancer-associated fibroblasts in breast cancer progression

In our previous studies we showed that in breast cancer podoplanin-positive cancer-associated fibroblasts (CAFs) correlated positively with tumor size, grade of malignancy, lymph node metastasis, lymphovascular invasion and poor patients' outcome. Therefore, the present study was undertaken to determine whether podoplanin expressed by fibroblasts can affect malignancy-associated properties of breast cancer cells. We found that migratory and invasive properties of breast cancer cells were not affected by the presence of podoplanin on the surface of human fibroblasts. However, we found that ectopic expression of podoplanin greatly increases the migration of such fibroblasts. Our experimental data were supported by clinical studies. When invasive ductal carcinoma (IDC) and ductal carcinoma in situ (DCIS) were analyzed by immunochemistry according to the presence of podoplanin-expressing CAFs, the numbers of CAFs with high expression of this glycoprotein were significantly higher in IDC than in DCIS cases. The present study also revealed, for the first time, that such podoplanin-mediated effects can influence angiogenesis. When human endothelial cells (EC) were co-cultured with podoplanin-rich fibroblasts the EC capillary-like network was characterized by significantly lower numbers of nodes and meshes than in co-cultures of endothelial cells with podoplanin-negative fibroblasts. The question

remains as to how our experimental data can be correlated with previous clinical data showing an association between the presence of podoplanin-positive CAFs and the progression of breast cancer. We propose that expression of podoplanin by fibroblasts facilitates their movement into the tumor stroma, which creates a favorable microenvironment for tumor progression by increasing the number of CAFs, which produce numerous factors affecting proliferation, survival and invasion of cancer cells. In accordance with this, the present study revealed, for the first time, that such podoplanin-mediated effects can influence angiogenesis and participate in its pathological properties in the tumor context.

DEPARTMENT OF ANTHROPOLOGY
Head: Professor Sławomir Kozieł

Post-migration adaptation and age at menarche in the second generation of migrants

Age at menarche is one of the most important measures of sexual maturation in girls. Since it has a high level of ecosensitivity, early environmental stress may trigger early puberty. One of these stress factors may be parental stress caused by the change of living conditions related to migration and adaptation to the new environment. Therefore, the aim of this study was to investigate the relationship between parental migration status and the timing of sexual maturity in the second generation, i.e. migrants' daughters. Data were collected during the 2nd Polish Anthropological Nationwide Field Investigation carried out in 1966-1969. The results show that age at menarche was accelerated in girls from low-socioeconomic-status (SES) migrant families in comparison to low-SES non-migrant families. This study provides new biosocial evidence on the impact of the parental long-lasting post-migration adaptation on the timing of maturity in the second generation of migrants.

Controlling behaviors and mate value discrepancy in women's romantic relationships

In behavioral ecology, controlling behaviors (CBs) are evolutionary strategies aiming to protect parental and relationship investments (P&RI) and reduce the risk of cuckoldry. Different forms of CBs are used by males and females. Similarly, from an evolutionary perspective, an individual's mate value (MV) describes all pheno- and genotypic aspects that promote successful reproduction. The rationale of the study was to analyze women's opinions and determine whether MV discrepancy between a woman and her heterosexual partner is related to the intensity of CBs used by both partners. Two evolutionary-based hypotheses were tested: i) partners of lower MV would more intensely control their higher MV partners; ii) the intensity of CBs would be higher in longer relationships (i.e. those with higher P&RI). The results showed that the highest intensity of CBs performed by both partners was in couples where a woman assessed her own MV as higher than her

partner's MV. This confirms the evolutionary hypothesis for men (a lower MV man controlled a higher MV partner) but not for women. More intense control of a lower MV partner performed by high MV women may result from social factors which probably mask biological effects. Higher intensity of CBs was also observed in longer relationships. This result is in line with the second hypothesis and supports the idea that CBs may operate as a behavioral strategy against squandering of high P&RI. The results of the study were published in the peer-reviewed journal *Acta Ethologica*.

Comparison of the disease structures observed in historical populations based on an analysis of skeletal paleopathological material unearthed from the medieval cemetery in Byczyna (11th–15th century) and the early modern (17th–19th century) cemetery at Czysty Square, Wroclaw

The aim of my study was to determine the prevalence of various diseases in populations that lived in medieval (11th–15th) Byczyna and early modern Wroclaw (17th–19th century) in the context of living conditions. I brought the results together to estimate the value of the relative similarity measure of these two structures. The calculated value indicates that in terms of the prevalence of different types of diseases, the two populations are only 4% similar. The occurrence of this difference was affected mainly by two factors: in the urban environment of early modern Wroclaw the incidence of infectious diseases (especially tuberculosis and syphilis) has significantly increased, as well as the incidence of rickets, which is an example of a deficiency disease. In the small-town environment of Byczyna, traumatic injuries and degenerative diseases were the most frequent, which could be possibly associated with a high activity level of the residents of this historical town. Taking into account the occurrence of these major differences, the research should be continued in order to find more detailed relationships between the health status and the structure of diseases of historical populations in the context of differentiated environmental conditions.

DEPARTMENT OF MICROBIOLOGY

Laboratory of Molecular Biology of Microorganisms
Head: Professor Anna Pawlik, Ph.D.

Replication of bacterial chromosomes
Polyketide synthesis and its regulation in Streptomyces

The research activity of the Laboratory of Molecular Biology of Microorganisms (LMBM) is focused on two scientific issues: i/ replication of bacterial chromosomes, and ii/ polyketide synthesis in *Streptomyces*.

i/ DNA replication is an important event of the bacterial cell cycle. The decision to initiate DNA replication is crucial for cell cycle progression, and depends both on intracellular and environmental signals. In LMBM we are interested in mechanisms of the initiation and regulation of bacterial

chromosome replication, with a special emphasis on the characterization of the key factors engaged in initiation complex (orisome) formation, namely DnaA – the initiator protein, and *oriC* – the origin of chromosome replication. We also aim at the identification and functional analysis of regulators, which control replication and coordinate the initiation with the cell cycle. We are especially interested in orisome formation in Epsilonproteobacteria, which comprise one of the classes within the phylum Proteobacteria. Epsilonproteobacteria are globally spread Gram-negative bacteria that inhabit a wide variety of ecological niches including water reservoirs, sewage, oil-field communities and deep-sea hydrothermal vents. Many of the known Epsilonproteobacteria are obligate or facultative human and/or animal pathogens including *H. pylori* – one of the most studied human-associated bacteria, causing gastric ulcer or gastric cancer. A few other Epsilonproteobacteria are assumed to be emerging human pathogens. Recently we identified and characterized *oriC*s in three epsilonproteobacterial species: pathogenic *Arcobacter butzleri*, symbiotic *Wolinella succinogenes*, and free-living *Sulfurimonas denitrificans.* Based on these data and thus far obtained results on the *H. pylori* orisome, we propose that *oriC*s typically co-localize with *ruvC-dnaA-dnaN*in Epsilonproteobacteria, with the exception of Helicobacteraceae species. The clusters of DnaA boxes localize upstream (*oriC1*) and downstream (*oriC2*) of *dnaA*, and they likely constitute bipartite origins. Unlike the DnaA box pattern, which is not conserved in Epsilonproteobacterial *oriC*s, the consensus DnaA box sequences and the mode of DnaA-DnaA box interactions are common to the class. Our results will facilitate identification of *oriC*s and subsequent identification of factors which regulate chromosome replication in other Epsilonproteobacteria. Since replication is controlled at the initiation step, it will help to better characterize the life cycles of these species, many of which are considered as emerging pathogens. The results will be beneficial for general knowledge concerning initiation of bacterial chromosome replication.

ii/ Polyketides are a large class of bioactive compounds with extremely diverse structures and functions. They are synthesized as secondary metabolites by giant multienzyme complexes – polyketide synthases. Our work is focused on the polyketide synthase Cpk from *S. coelicolor* A3(2), which is responsible for the synthesis of a yellow pigment, coelimycin. Expression of *cpk* genes is tightly controlled by regulatory proteins encoded by the genes within the *cpk* cluster and probably by several pleiotropic regulators connected with regulation of secondary metabolite production as well as *Streptomyces* morphological differentiation. We are interested in deciphering the regulatory circuits governing the synthesis of coelimycin as well as in the discovery of its biological activity.

Laboratory of Signal Transduction Molecules
Acting Head: Professor Jakub Siednienko, Ph.D.

Studies on proteins and signaling pathways involved in activation of proinflammatory transcription factors and the response to hypoxia

The innate immune response is a universal mechanism of defense against invading pathogens. Toll-like receptors (TLRs), being a part of this first line of defense, are responsible for detecting a wide variety of microorganisms including bacteria, viruses and fungi, and, in some cases, a number of host molecules – the breakdown products from ruptured cells and tissues. When a TLR receptor recognizes a disease-causing agent, it initiates an immune response, thus facilitating removal of the pathogen.

TLR3, TLR7/8 and TLR9 are receptors that mainly recognize viral components. TLR7 recognizes single-stranded RNA, a genetic material of some viruses such as the influenza virus. Despite the differences in ligand specificity the effect of activation of both TLR-dependent pathways is the production of antiviral proteins – type I interferons, cytokines and chemokines. So far only a few proteins have been characterized as essential for signal transduction from the activated receptor to the effector transcription factors.

On the basis of our experimental results we identified new Mal-dependent mechanisms that regulate the immune response against viral infections especially in the TLR7 and TLR9 context. At the molecular level we identified key kinases, ERK 1/2, and transcription factors, p105, c-Rel and IRF7, that are involved in the immune response regulated by Mal in TLR 7 and TLR9 signaling pathways.

DEPARTMENT OF TUMOR IMMUNOLOGY

Laboratory of Molecular and Cellular Immunology
Head: Professor Małgorzata Cebrat, Ph.D.

The role of NWC protein in murine spermatogenesis

The *NWC* gene, which was discovered by our team, is strongly conserved among vertebrates and invertebrates. Despite the ubiquitous presence of the *NWC* transcript in several murine tissues and cell types (except for mature T and B lymphocytes), the NWC protein is detected only in the testis. To gain insight into the function of NWC protein we have generated NWC-knockout mice (NWC-KO). Compared to the wild-type (WT) mice, NWC-KO mice did not reveal any apparent phenotypic changes; however, due to the presence of NWC in the testis, we decided to investigate in detail possible deficiencies in male fertility of the NWC-KO mice.

First, we determined NWC distribution in seminiferous epithelial cells. We found that NWC is present in preleptotene, leptotene, zygotene, pachytene spermatocytes and in round spermatids up to steps 11-12. We did not detect NWC in sperm cells recovered from epididymis and in Sertoli cells. We did not find differences between NWC-KO and WT in number of sperm cells recovered from epididymides or in testis weight. Hematoxylin-eosin staining did not reveal any changes in testis structure.

In order to fertilize oocytes, sperm cells need to undergo capacitation – a process of functional reprogramming manifested at biochemical and physiological levels. Eventually, capacitation enables the sperm cells to undergo acrosome reaction. We have shown that a population of acrosome reacted sperm cells recovered from epididymis of NWC-KO males was smaller than a population of control WT sperm cells.

Standard, unrestricted breeding protocols applied for reproducing mice in laboratory conditions do not reflect the complex mating behavior of these animals. Female mice are polyandrous as in the natural environment during estrus they can mate with multiple males, which leads to simultaneous presence of sperm cells from different individuals in a single female reproductive tract and subsequent sperm cell competition. It is important to stress that taking into account sperm cell competition during analysis of fertility might be crucial for finding a critical role of particular molecules in male fertility. We have shown that NWC-KO sperm cells have lower chances to fertilize oocytes when we experimentally induced competition with WT sperm cells in the female reproductive tract. If a WT male was given access to the female first, about 90% of embryos had the WT genotype, indicating that WT sperm cells had won the competition with NWC-KO sperm cells. When the accession order was reversed, NWC-KO males failed to reproduce similar success and were even unable to fertilize a higher number of oocytes than WT males. This result indicates that NWC-KO sperm cells might be functionally impaired.

Verifying our previous discoveries, we have also found, using bimolecular fluorescence complementation, that NWC protein interacts with IFT122 (IFT – intraflagellar transport) protein.

To summarize, we conclude that NWC-KO sperm cells are functionally impaired, as indicated in sequential mating experiment when they compete with WT sperm cells to fertilize oocytes. This might be a result of a weaker response of NWC-KO sperm to factors triggering acrosome reaction. We postulate that the effects of NWC deletion is mediated by NWC engagement in intraflagellar (IFT) and/or intramanchette (IMT) transport systems.

Laboratory of Tumor Immunology
Head: Professor Arkadiusz Miążek, Ph.D.

Effect of anti-GITR antibody treatment on increase in diversity of regulatory T cells infiltrating B16 melanoma tumors

Cross-linking of glucocorticoid-induced TNF family related receptor (GITR) with agonist antibodies restores cancer immunity by enhancing effector T cell (Teff) responses while interfering with intra-tumor regulatory T cell (Treg) stability and/or accumulation. However, the direction of the T cell receptor (TCR) repertoire changes associated with anti-GITR antibody infusion and their significance for tumor eradication is unclear. Here, we used a transgenic mouse model (TCRmini) where T cells express a naturally generated but restricted TCR repertoire to trace the fate of individual T cells specific for B16 melanoma in tumor-bearing mice, treated or not treated with the anti-GITR monoclonal antibody DTA-1. Analysis of TCRs from CD4+ cells sorted from these mice revealed that the TCR repertoire of dominant Teff clones locally skewed toward tumor antigens remained roughly similar in treated and non-treated mice. In contrast, both the tumor-associated and peripheral TCR repertoire of Tregs, which was in the majority distinct from that of Teffs, underwent DTA-1 mediated remodeling characterized by depletion of dominant clones and the emergence of more diverse, low-frequency clones bearing increased numbers of TCRs shared with Teffs. We conclude that the single dose DTA-1 infusion entails a double-edged sword effect by eliminating activated Tregs engaged in the initial maintenance of a tolerogenic niche for tumor growth on one hand, but on the other hand by favoring progressive tumor replenishment by a Treg pool fitted with TCRs able to better compete with Teffs for recognition of common tumor antigens.

DEPARTMENT OF EXPERIMENTAL THERAPY
Head: Professor Michał Zimecki, Ph.D.

Laboratory of Immunobiology
Head: Professor Michał Zimecki, Ph.D.

Studies on immunosuppressive properties of selected azaphenothiazines

Several azaphenothiazines, previously selected as strongly antiproliferative agents in *in vitro* models, were subjected to evaluation for their potential immunosuppressive effects in the model of delayed type hypersensitivity (DTH) to ovalbumin (OVA) in BALB/c mice and in foot pad inflammation induced by carrageenan in CBA mice. Among the compounds, significantly suppressive activities in both models were exhibited only by compound **5** (6-chloroethyl-ureidoethyldiquino[3,2-b;2',3'-e][1,4]thiazine) and compound **4** (6-acetylaminobutyl-9-chloroquino [3,2-b]benzo[1,4]thiazine). Molecular studies revealed that compound **5** blocked expression of caspase 3 and strongly inhibited expression of caspases 8 and 9 in Jurkat cells. Moreover,

compound **5**, but not **4**, induced cell apoptosis in several tumor cell lines. We also investigated the efficacy of topically applied compounds **4** and **5** in amelioration of inflammatory symptoms of contact sensitivity (CS) to oxazolone in mice, in relation to Protopic, the reference drug. The results demonstrated efficient therapeutic properties of both compounds applied in the form of an ointment at the time of maximal manifestation of the cutaneous inflammatory reaction.

Studies on immunoregulatory properties of isoxazoles

Isoxazoles are an important class of compounds of potential therapeutic value. The aim of this study was to determine immunotropic effects of 5-amino-3-methyl-4-isoxazolecarboxylic acid hydrazide derivatives on spontaneous and mitogen-induced lymphocyte proliferation in young and old BALB/c mice, cytokine production by peritoneal cells as well as the possible mechanism of action in a model of Jurkat cells. The mice were used as donors of the cells from thymus, spleen, mesenteric lymph nodes, and peritoneal cavity. 5-amino-3-methyl-4-isoxazolecarboxylic acid hydrazide, a parental compound, **01K** (4-phenyl-1-(5-amino-3-methylisoxazole-4-carbonyl)-thiosemicarbazide), and **06K** (4-(4-chlorophenyl)-1-(5-amino-3-methylisoxazole-4-carbonyl)-thiosemicarbazide) exhibited regulatory activity in the proliferation tests. Prevailing stimulatory activity of the hydrazide and inhibitory activity of **01K** and **06K** was observed. Those effects were connected with different influence of the compounds on signaling proteins' expression in Jurkat cells. The regulatory effects of the compounds on interleukin 1 beta (IL-1β) production were more profound than those on tumor necrosis factor alpha (TNF-α). Differences in the compound activity in young versus old mice were mainly restricted to 01K. Leflunomide was used as a reference drug. Immunoregulatory properties of compound **06K** were also tested *in vivo* for its effects on humoral and cellular immune response, carrageenan inflammatory reaction and composition of lymphocyte subsets in non-immunized mice. The compound administered before or after immunization with sheep erythrocytes (sheep red blood cell (SRBC)) elevated the number of plaque-forming cells (PFC), and this effect was stronger at lower doses. Although total hemagglutinin titers to SRBC decreased upon post-immunization treatment, the IgG titer increased. In the DTH model to OVA, the compound, applied intraperitoneally before an eliciting dose of an antigen but not before immunization, inhibited the magnitude of a cutaneous reaction. Furthermore, **06K** significantly diminished carrageenan-induced foot pad inflammation when administered 1 h before carrageenan. The compound, administered intraperitoneally to naïve mice, elicited changes in weight, cell number in lymphoid organs and content of lymphocyte subsets, depending on the dose and number of applications. Phenotypic changes included increased turnover of thymocytes, changes in B-cell distribution in spleens and lymph nodes, increased percentage of CD8+ cells and regulatory CD4+

CD25+ Foxp3+ T cells. In summary, the immunoregulatory properties of **06K** involved mobilization of lymphopoiesis and generation of regulatory T cells.

Laboratory of Immunopathology
Head: Professor Irena Frydecka, M.D, Ph.D.

Studies on the mechanisms of immune deficiency in neoplastic and autoimmune diseases

Association of genetic variations within CTLA-4, CD28, ICOS and TPO genes with Graves' disease in its phenotype

Graves' disease (GD), an autoimmune disease with heterogeneous symptoms including Graves' orbitopathy, has a combined genetic/environmental background, where several genes may by involved in the pathogenesis, including those encoding co-stimulatory molecules CD28/CTLA-4/ICOS and thyroid peroxidase (TPO) – a key enzyme of thyroid function, having a pivotal role in the synthesis of thyroid hormones. We selected functional polymorphisms located within genes encoding co-stimulatory molecules: *CD28*c.17+3T>C (rs3116496), *CTLA-4*g.319C>T (rs5742909), *CTLA-4*c.49A>G (rs231775), *CTLA-4*g.*642AT(8_33), CT60 (*CTLA-4*g.*6230G>A, rs3087243), Jo31 (*CTLA-4*g.*10223G>T, rs11571302), *ICOS*c.1554+4GT(8_15) and the most significantly associated SNP in the TPO region identified in the genome-wide association study: rs11675434. Genetic variation within genes encoding co-stimulatory molecules was studied in 561 Polish Caucasians, including 172 unrelated Graves' disease patients, whereas SNP in the TPO region was studied in a group of 1231 well-characterized patients with GD (1043 adults and 188 children) and 1130 healthy controls.

*CD28*c.17+3T>C[T]/*CTLA-4*g.319C>T[C]/*CTLA-4*c.49A>G[G]/*CTLA-4*g.*642AT (8_33)($AT_{16–21}$)/CT60[G]/Jo31[G]/*ICOS*c.1554+4GT(8_15)(m) and TCA($AT_{<16}$)GT(m) haplotypes increased the risk of Graves' disease, especially in males, as well as overall Graves' orbitopathy development with severe outcome. TCG($AT_{16–21}$)GG(l) haplotype increased the risk of Graves' disease and reduced the chance of successful medical treatment. Although this haplotype was mainly observed in patients without signs of Graves' orbitopathy, if Graves' orbitopathy developed it favored a Graves' orbitopathy outcome. Haplotype TCA($AT_{>21}$)GT(m) increased Graves' disease risk in women and, in all patients, was linked to Graves' disease without Graves' orbitopathy. TCG($AT_{<16}$)GG(m) haplotype was predominantly observed in patients without Graves' orbitopathy, whereas TCA($AT_{16–21}$)GG(m) was absent in those patients. TCA($AT_{16–21}$)GG(m) occurred in patients with a mild Graves' orbitopathy outcome. The marker *CTLA-4*g.*642AT(8_33) was the only independent Graves' disease risk factor, whereas CT60 was an independent factor for disease progression.

At a univariate level, sporadic Graves' disease was related to presence of *CTLA-4*c.49A>G[A] and the rare *CTLA-4*g.319C>T[T] allele variant. Familial background of the disease was associated with *CTLA-4*g.*642AT(8_33)[$AT_{>21}$]/[$AT_{>21}$] genotype.

We found that the T allele of rs11675434 was significantly more frequent in GD patients with than without GO. Further analyses performed in subgroups of patients showed that the association with GO was significant in adult patients with age of GD onset ~45 years, but not in children and adolescents or adult patients with earlier onset of the disease. Moreover, a strong association with GO was present in males, whereas it was absent in females.

Taken together, our results showed that *CD28/CTLA-4/ICOS* loci as well as the TPO region may confer inherited susceptibility to Graves' disease and its outcome – severity and activity of Graves' orbitopathy. Moreover, our results showed that apart from genetic markers, also environmental parameter such as gender and familial autoimmune thyroid history should be taken into account when a patient's genetic background is analyzed.

Laboratory of Reproductive Immunology
Head: Professor Anna Chełmońska-Soyta, Ph.D., V.D.

Immunological mechanisms associated with reproductive processes in health and disease

Expression of IL-24 in T cells and B regulatory females and males

IL-24 is a cytokine belonging to the IL-10 cytokine family. It is produced by T and B lymphocytes, macrophages, cancer cells and trophoblast cells. In mice it has been shown that it is produced by Th2 lymphocytes differentiated by IL-4. It has immunomodulatory properties exerted by inhibiting the proliferation of cytotoxic T cells. Moreover, it shows pro-apoptotic activities in cancer cells and inhibits proliferation of trophoblast cells. In previous studies, we have demonstrated the presence of IL-24 in CD4+ T cells in the spleen of mice during pre-implantation pregnancy after embryo transplantation.

The aim of this study was to test whether IL-24 is produced by lymphocyte subpopulations with regulatory properties, i.e. Treg cells (CD4+ CD25+ FOXP3+) and Breg (CD19+ CD1d+ CD5+ B220 +) in the central and peripheral lymphoid organs in CBA/J mice. Studies were carried out in male CBA/J mice at the age of 5 weeks. IL-24 was detected within the cell after stimulation with PMA and ionomycin. The presence of IL-24 was demonstrated in 76.8 +/- 11.97% of double positive thymocytes CD8b CD8a++ FOXP3+ CD4+ and 95.85+/- 1.69% of double negative thymocytes CD8b-CD8a-CD4+ FOXP3+ ($p < 0.005$). In regulatory T cells (CD4+ CD25+ FOXP3+) at the periphery the presence of IL-24 was confirmed in more than 70% of cells in the spleen, lymph nodes, and over 60% of the cells in the blood. Similarly, in B regulatory cells

(CD19+ CD220+ CD5+ Cd1high) the presence of IL-24 was confirmed in over 70% of the cells in the spleen, lymph nodes and blood. In conclusion: the ubiquitous presence of IL-24 in regulatory lymphocytes may be used as an additional phenotype marker of these cells.

DEPARTMENT OF IMMUNOLOGY OF INFECTIOUS DISEASES
Head: Professor Andrzej Gamian, Ph.D.

Laboratory of Medical Microbiology
Head: Professor Andrzej Gamian, Ph.D.

Studies on the pathogenesis of some diseases of bacterial etiology and the role of bacterial surface glycoconjugates and protein antigens in the immune response, as well as studies on probiotic glycoconjugates

Studies performed in our laboratory concern mechanisms of pathogenicity of diseases with bacterial etiology, the role of bacterial glycoconjugates and proteins in the immune processes, and the structure and functions of bacterial exopolysaccharides, including those from probiotics. Studies on the OmpC protein recognized by umbilical cord serum revealed that cyclic hexapeptide but not linear mimics the unique conformational epitope of OmpC protein, exposed on the cell surface of *Shigella flexneri*. Such a cyclic peptide is a proper antigen for construction of a conjugate vaccine, potent to induce protective immunity. Interesting results have been obtained from a survey on the global phylogeography and evolutionary history of *Shigella dysenteriae* type 1. The strains kept in our Polish Collection of Microorganisms (PCM) have been identified among world epidemics of dysentery. Regarding the studies on advanced glycation end-products, a mouse monoclonal antibody has been obtained, of IgE class. In order to investigate whether this antigen is present in different animals, its distribution in various species was investigated with immunohistochemical experiments. This epitope was detected in tissues of rat, rabbit, swine, sheep, hen, frog, fish and snail, which indicates its common occurrence. Determination of the level of this antigen in different clinical cases will allow for better understanding of the biology of these advanced glycation end-products and their significance in pathology. Other studies indicate that major glycolipids of *Lactobacillus johnsonii* may serve as new biomarkers for inflammatory bowel disease diagnostics. Identification of *Bifidobacterium* and *Lactobacillus* proteins immunoreactive with human serum, together with umbilical cord serum and revealing their immunogenic properties, is a milestone in understanding the biology of these microorganisms regarding their protective activity to pathogens and commensals. In this context there is important observation on the reactivity of one of the polysaccharides from *Lactobacillus* with human adult sera, while it is not recognized by umbilical cord serum. Exopolysaccharides from probiotics present a potential for immune system modulation.

Laboratory of Virology
Head: Professor Egbert Piasecki, Ph.D.

Study on nonspecific immunity in viral infections

Natural killer cells play an important role as effectors of innate immunity and regulators of adaptive immunity. They are important elements of the innate response to viral infections, which they detect using human leukocyte antigen (HLA) class I-binding receptors. The most polymorphic of these are killer cell immunoglobulin-like receptors (KIRs), which exist as two basic isotypes, activating or inhibitory receptors, and are encoded by genes distributed differently in unrelated individuals. We searched for links between selected clinical data (including HCV viremia, liver enzyme levels and liver histology parameters) and the presence of genes encoding these receptors and their ligands in hepatitis C virus-infected individuals subjected to pegylated interferon-a and ribavirin therapy. Genomic DNA samples from two hundred and ninety-two chronically infected patients were typed by polymerase chain reaction for the presence or absence of genes for KIRs and their ligands, class I HLA molecules, and clinical data of the patients were collected. Our results suggest the importance of clinical parameters and the contribution of KIR and HLA genes to the course of hepatitis C virus infection and the response to therapy. The study revealed that levels of liver enzymes before therapy were about 30% higher in patients who possessed a variant KIR2DS4 gene with 22-base pair deletion. The decrease of ALT activity after treatment was higher in HLA-C C2-positive than negative individuals. Additionally, patients demonstrated an early virologic response to the therapy if the time lag before treatment was short, particularly in women. The results were published in *Archivum Immunology and Experimental Therapy*, 2016; 64: 65-73.

Killer cell immunoglobulin-like receptors (KIR) are the most polymorphic receptors of natural killer (NK) cells. Their activity diversifies the functions of NK cells in the antiviral immune response, so the presence of certain KIR may affect transmission of HIV-1. The aim of the study was to evaluate the influence of KIR genes on the susceptibility to HIV-1 infection in the Polish population depending on the route of exposure. We determined the frequencies of activating (2DS1, 2DS2, 2DS3, 2DS4f, 2DS4del, 2DS5, 3DS1) and inhibitory (2DL1, 2DL2, 2DL3, 2DL5, 3DL1) KIRs in HIV-1-positive patients (n= 459), individuals exposed to HIV-1 but uninfected (EU, n=118) and in uninfected, healthy blood donors (BD, n=98). Analysis was performed using stepwise logistic regression. Apart from KIRs, CCR5-Δ32, and CCR2-64I, alleles were also analyzed, as we knew or suspected that these features could affect susceptibility to HIV infection. The regression confirmed the protective effect of CCR5-Δ32 (OR=0.25, p=0.006) and CCR2-64I (OR = 0.59, p = 0.032) against HIV infection. Among KIR genes, 2DL3 was found to be a protective factor (OR=0.30, p=0.015). A similar effect was seen for 3DS1 but only in intravenous

drug users (IDUs) (OR=0.30, p=0.019), not in sexually exposed people. 2DL5 was found to be a factor facilitating HIV infection (OR=2.13, p=0.013). A similar effect was observed for 2DL2 but only in females (OR=2.15, p=0.040), and 2DS1 in IDUs (OR=3.03, p=0.022). Our results suggest a beneficial role of KIR3DS1 and 2DL3 supporting resistance to HIV infection and harmful effect of 2DS1, 2DL5, and 2DL2 genes promoting HIV acquisition. The results were published in *Immunogenetics*, 2016; 68: 327-337.

Complex genetic diagnosis of seronegative (HIV-) long-term partner of female patient with AIDS C3 in the context of HIV transmission – case report. HIV infection was excluded in a 47-year-old man, a long-term sexual partner of a female patient diagnosed with AIDS C3. The risk of HIV infection was estimated as high. We focused on the genetic diagnosis of the serodiscordant couple. We determined the presence of CCR5-Δ32, CCR2-64I, HLA-B, killer cell immunoglobulin-like receptor (KIR) and their ligand genes and human endogenous retroviruses K113 and K115. Genotyping was performed using PCR methods. Analysis of the partner's genotype revealed the presence of CCR5-Δ32/Δ32 and KIR genes encoding activating receptors (KIR2DS1, 2DS5, 3DS1), features associated with reduced risk of HIV transmission. Similarities in the patient's and her partner's HLA (HLA-B*51) and similar inhibitory KIR repertoire and their ligands (KIR2DL1+HLA-C2, KIR2DL3+HLA-C1, KIR3DL1+HLA-B Bw4-80Ile) could favor the transmission of the virus. Genetic diagnosis is not routinely recommended. Observation of exposed and uninfected individuals would allow implementation of new knowledge into more effective care. The results were published in *HIV & AIDS Review*, 2016; 15: 97-100.

DEPARTMENT OF IMMUNOCHEMISTRY
Head: Professor Czesław Ługowski, Ph.D.

Laboratory of General Immunochemistry
Head: Professor Maria Janusz, Ph.D.

Neuroprotective properties of the naturally derived polypeptide complexes
Yolkin effect on ERK1/2 kinase activation and protection against toxic effect of free radicals

Egg yolk proteins are considered a rich source of nutrients. Among them an important role is played by yolkin – a polypeptide complex with properties similar to colostrum-derived proline-rich polypeptide complex PRP/Colostrinin. Yolkin modulates both pro- and anti-inflammatory cytokine secretion. The effect of Yolkin on improvement of locomotor function and exploratory behavior, preventing their decline, and the functioning of episodic and spatial memory in aging rats was observed. The neuroprotective effect of Yolkin is

connected with its positive effect on cell viability and brain-derived neurotrophic factor (BDNF) secretion.

One of the primary causes of neurodegeneration of nerve cells is oxidative stress. The endogenous system (both enzymatic and low molecular antioxidants) protects cells against toxic effects of free oxygen radicals (ROS). When ROS secretion inside cells is elevated in the presence of Aβ42 or tau protein the structure and function of nerve cells are disturbed. In oxidative stress condition protein kinases ERK1/2 are induced. Their sustained activation can cause mitochondrial destruction, Bc1 protein activation and, as an end effect, neuronal apoptosis. To consider the mechanism of Yolkin action in neurodegenerative processes it was interesting to assess its modulatory effect on ERK1/2 activation and regulation of ROS intracellular secretion.

A Yolkin preparation was tested for its purity in SDS PAGE and its activity was checked as ability to induce IL6 secretion in whole blood cells. As model cells the neuronal rat cell line PC12 was used. It was found that Yolkin does not induce free radical species. However, in oxidative stress conditions (induced with the use of 75 μM H_2O_2) Yolkin in a dose-dependent manner reduces intracellular ROS accumulation (20–30%). Moreover, it was observed (by Western blotting) that in oxidative stress conditions Yolkin inhibits sustained activation of ERK1/2 kinases.

The results obtained indicate that Yolkin neuroprotective properties are connected with reduction of the ROS level.

Laboratory of Glycoconjugate Immunochemistry
Head: Professor Hubert Krotkiewski, Ph.D.

Evaluation of relationship between polymorphisms in human A4GALT gene and the number of P1PK blood group antigens on erythrocytes

The P1PK blood group system consists of three glycosphingolipid antigens: P^k (Gb3, CD77), P1 and NOR. The P^k antigen is expressed on most human red blood cells (except those of p phenotype), whereas P1 is present only in a fraction with ethnicity-specific frequency, thus underlying two common phenotypes: P_1, if the P1 antigen is present, and P_2, if the P1 antigen is absent. The molecular mechanism for the formation of P_1/P_2 blood groups remains not fully elucidated. We sequenced the promoter region of the *A4GALT* gene from 85 individuals of different PIPK status, and evaluated the *A4GALT* transcript level and the P1PK antigen number on erythrocytes. Conclusions are as follows:

1. The best correlation between genotype and P1PK number was found for the SNP rs5751348.
2. The number of P1PK antigens on erythrocytes from individuals of different genotypes is as follows: $P^1P^1 > P^{1NOR}P^1 > P^1P^2 > P^{1NOR}P^2 > P^2P^2$.
3. Presence of NOR antigen causes a decrease of levels of other P1PK antigens.
4. There is no correlation between the LDL/HDL cholesterol level and the number of P1PK antigens on erythrocytes.

Distribution of Duffy blood group antigens in Polish population

Duffy genotyping of 596 unrelated donor blood samples was investigated by high-resolution melting (HRM) analysis. It was found that phenotype Fy(a+b+), defined by the heterozygous genotypes *FY*A/FY*B* (33%) and *FY*A/FY*B298A* (13%), was the most common in the Polish population (~46%), followed by Fy(a-b+), ~29%, determined by the genotypes raised from the *FY*B* allele and all its variants. Phenotype Fy(a+b-) occurred with a frequency of 24% and was defined by the following genotypes: *FY*A/FY*A* (21%), *FY*A/FY*X* (2.8%) and *FY*A/FY*B-33* (0.3%). Among the Polish population the frequencies of *FY*A*, *FY*B*, and *FY*B298A* alleles were 45.7%, 36% and 15.5%, respectively. The alleles *FY*B298A* and *FY*B*, combined together, represented higher frequency (51%) than *FY*A*. Alleles *FY*X* and *FY*B-33* had low frequencies of 2.51% and 0.25%, respectively. To our knowledge the present study is the first to report the genotypes and allele frequencies of the *FY* gene in the Polish population.

Laboratory of Microbial Immunochemistry and Vaccines
Head: Professor Czesław Ługowski, Ph.D.

Biochemical characteristics of macromolecules involved in immunological processes. Immunochemical studies of bacterial endotoxins

The complement system is a part of innate immunity and represents the first line of defense against infections. Its activation leads to initiation of the inflammatory response followed by death of the pathogen. There are three pathways of complement activation: classical, alternative, and lectin-based. The lectin pathway in humans is initiated by certain collectins, such as mannose-binding lectin (MBL), collectin-10 (CL-10), collectin-11 (CL-11), and all known ficolins, including ficolin-1 (M-ficolin, p35-related protein), ficolin-2 (L-ficolin) and ficolin-3 (H-ficolin). Despite recombinant protein technology development, proteins isolated from natural sources remain important for structure and activity determination. Ficolins represent a class of proteins that are

difficult to isolate. To date, three methods for purifying ficolin-3 from plasma/serum have been proposed, defined by the most critical step: (i) hydroxyapatite absorption chromatography, (ii) N-acetylated human serum albumin affinity chromatography, and (iii) anti-ficolin-3 monoclonal antibody-based affinity chromatography. We present a new protocol for purifying ficolin-3 complexes from human plasma that is based on an exclusive ligand: the O-specific polysaccharide of *Hafnia alvei* PCM 1200 LPS (O-PS 1200). The protocol includes (i) poly(ethylene glycol) precipitation; (ii) yeast and L-fucose incubation, for depletion of mannose-binding lectin; (iii) affinity chromatography using O-PS 1200-Sepharose; and (iv) size-exclusion chromatography. Application of this protocol yielded on average 2.2 mg of ficolin-3 preparation free of mannose-binding lectin (MBL), ficolin-1 and -2 from 500 ml of plasma. The protein was complexed with MBL-associated serine proteases (MASPs) and was able to activate the complement *in vitro*. In-process monitoring of MBL, ficolins, and total protein content revealed the presence of difficult-to-remove immunoglobulin G, M and A, to some extent in agreement with recent findings suggesting crosstalk between IgG and ficolin-3. We demonstrated that recombinant ficolin-3 interacts with IgG and IgM in a concentration-dependent manner. Although this association does not appear to influence ficolin-3-ligand interactions *in vitro*, it may have numerous consequences *in vivo*. Thus our purification procedure provides Ig-ficolin-3/MASP complexes that might be useful for gaining further insight into the crosstalk and biological activity of ficolin-3.

Publication - 2016

Articles published in the journals from Thomson Reuters Master Journal List:

1. Antoszczyk M, Maj E, Kleczewska N, Wietrzyk J, Celewicz L, Huczyński A.: Differences in Antiproliferative Activity between Salinomycin-AZT Conjugates Obtained via 'Click' and Esterification Reactions. *Med Chem.*, 2016; 12 (8), doi: 10.2174/ 1573406412666160823165522 **IF – 1,458 (20 pkt. Min.)**
2. Artym J, Kocięba M, Zaczyńska E, Kochanowska I, Zimecki M, Jeleń M, Morak-Młodawska B, Pluta K.: Selected azaphenothiazines inhibit delayed type hypersensitivity and carrageenan reaction in mice. *International Immunopharmacology*. 2016, 40, 265-268; doi:10.1016/ j.intimp.2016.09.005 **IF – 2,551 (25 pkt. Min.)**
3. Bączkowicz D, Falkowski K, Majorczyk E. J.: Assessment of relationships between joint motion quality and postural control in patients with chronic ankle joint instability. *Orthop Sports Phys Ther*, 2016, 4: 1-26 **IF - 2,550 (45 pkt. Min.)**
4. Bączkowicz D, Majorczyk E.: Joint motion quality in chondromalacia progression assessed by vibroacoustic signal analysis. PM & R: the journal of injury, function, and rehabilitation. 2016, pii: S1934-1482(16)30058-2. doi: 10.1016/j.pmrj.2016.03.012. **IF – 1,655 (35 pkt. Min.)**
5. Badowska-Rosłonek K, Ciesielska A, Świtalska M, Piskozub M, Peczyńska-Czoch W, Wietrzyk J, Kaczmarek Ł.: Synthesis and cytotoxic activity of new 5h-indolo[2,3-b]quinoline o-aminoglycosides. *Acta Pol Pharm*. 2016; 73(3):683-92. **IF – 0,877 (15 pkt. Min.)**

6. Bębenek E, Kadela-Tomanek M, Chrobak E, Wietrzyk J, Sadowska J, Boryczka S.: New acetylenic derivatives of betulin and betulone, synthesis and cytotoxic activity. *Med Chem Res*, 2016, doi:10.1007/s00044-016-1713-9. **IF – 1,436 (20 pkt. Min.)**
7. Bębenek E, Chrobak E, Wietrzyk J, Kadela M, Chrobak A, Kusz J, Książek M, Jastrzębska M, Boryczka S.: Synthesis, structure and cytotoxic activity of acetylenic derivatives of betulonic and betulinic acids, *J Mol Structure*, 2016, 1106: 210–219. **IF – 1,708 (20 pkt. Min.)**
8. Bentham J., Di Cesare M., Koziel S., Zuniga Cisneros J.: (NCD Risk Factor Collaboration): A century of trends in adult human height, *eLife*, 2016, 5(July): e134410. DOI: 10.7554/eLife.13410.001 **IF – 8,282 (45 pkt. Min.)**
9. Bienias K., Fiedorowicz A., Sadowska A., Prokopiuk S., Car H.: Regulation of sphingomyelin metabolism. *Pharmacol. Rep*., 2016, 68: 570-581. **IF - 2,251 (25 pkt. Min.)**
10. Biernat M, Cydzik M, Stefanowicz P, Kluczyk A, Artym J, Zimecki M, Szewczuk Z:. Cyclodimerizaton of immunosuppressive fragment of HLA-DR molecule. Design, synthesis and ESI-MS/MS analysis. *Journal of Peptide Science*, 2016, 22, 525-532; doi: 10.1002/psc.2898; **IF – 1,951 (20 pkt. Min.)**
11. Biskup I, Zaczynska E, Krauze-Baranowska M, Fecka I.: Evaluation of cytotoxicity of 5-*n*-alkylresorcinol homologs and fraction on mouse fibroblast cell line L929. *Eur Food Res Technol*, 2016. doi:10.1007/s00217-016-2827-5 **IF - 1,433 (25 pkt. Min.)**
12. Błachowicz O, Zwolińska K.: Rola genów kodujących immunoglobulinopodobne receptory komórek cytotoksycznych (KIR) oraz ich ligandy w podatności i przebiegu zakażenia HIV. *Post Hig Med Dosw*, (Online), 2016; 70: 1409-1423 **IF – 0,769 (15 pkt. Min.)**
13. Błażejczyk A, Switalska M, Chłopicki S, Marcinek A, Gebicki J, Nowak M, Nasulewicz-Goldeman A, Wietrzyk J.: 1-methylnicotinamide and its structural analog 1,4-dimethylpyridine for the prevention of cancer metastasis. *J Exp Clin Cancer Res*, 2016**,** 35**:**110, doi: 10.1186/s13046-016-0389-9 **IF – 4,357 (30 pkt. Min.)**
14. Bocian K., Borysowski J., Zarzycki M., Pacek M., Weber-Dąbrowska B., Machcińska M., Korczak-Kowalska G., Górski A.: The effects of T4 and A3/R bacteriophages on differentiation of human myeloid dendritic cells. *Frontiers in Microbiology*, 2016, vol 7, art. 1267 **IF – 4,165 (35 pkt. Min.)**
15. Bocian K., Borysowski J., Zarzycki M., Wierzbicki P., Kłosowska D., Weber-Dąbrowska B., Korczak-Kowalska G., Górski A.: LPS- activated monocytes are unresponsive to T4 phage and T4 – generated *Escherichia coli* lysate. *Frontiers in Microbiology*, 2016, vol 7, art. 1356 **IF – 4,165 (35 pkt. Min.)**
16. Bogunia-Kubik K., Wysoczańska B., Piątek D., Iwaszko M., Ciechomska M., Świerkot J.: Significance of polymorphism and expression of miR-146a and NFkB1 genetic variants in patients with rheumatoid arthritis. *Arch Immunol Ther Exp*, 2016, 64, suppl. 1, 131-136 **IF - 2,464 (25 pkt. Min.)**
17. Brzozowska E, Koba M, Śmietana M, Górska S, Janik M, Gamian A, Bock WJ.: Label-free Gram-negative bacteria detection using bacteriophage-adhesin-coated long-period gratings. *Biomed Opt Express*, 2016 Feb 9; 7(3):829-40. **IF – 3,344 (40 pkt. Min.)**
18. Burchacka E, Witkowska D.: The role of serine proteases in the pathogenesis of bacterial infections. [2]. *Post Hig Med Dosw*, (Online), 2016 Jun 30;70(0):678-94. **IF – 0,769 (15 pkt. Min.)**
19. Butrym A., Gebura K., Iwaszko M., Kuliczkowski K., Bogunia-Kubik K., Mazur G.: Dual role of the CXCL12 polymorphism in patients with chronic lymphocytic leukemia. *HLA (Immune Response Genetics)*, 2016; 87(6): 432-8. **IF – 2,046 (20 pkt. Min.)**

20. Butrym A., Łacina P., Rybka J., Chaszczewska-Markowska M., Mazur G., Bogunia-Kubik K.: Cereblon and IRF4 variants affect risk and response to treatment in multiple myeloma. *Arch Immunol Ther Exp***,** 2016**,** 64, suppl. 1 151-156 **IF - 2,464 (25 pkt. Min.)**
21. Chaszczewska-Markowska M., Sagan M., Bogunia-Kubik K.: The renin-angiotensin-aldosterone system (RAAS) - physiology and molecular mechanisms of functioning. *Post Hig Med Dosw,* (online), 2016; 70: 917-927. **IF - 0,769 (15 pkt. Min.)**
22. Ciechomska M., O'Reilly S., Przyborski S., Oakley F., Bogunia-Kubik K., van Laar JM.: Histone Demethylation and Toll-like Receptor 8-Dependent Cross-Talk in Monocytes Promotes Transdifferentiation of Fibroblasts in Systemic Sclerosis Via Fra-2. *Arthritis Rheumatol,* 2016; 68(6): 1493-504. **IF – 6,009 (45 pkt. Min.)**
23. Ciekot J, Goszczyński TM, Boratyński J.: Methods for preliminary determination of pemetrexed in macromolecular drug-carrier systems. *Acta Pharm*. 2016 Mar; 66(1):147-53. doi: 10.1515/acph-2016-0001. **IF – 1,212 (20 pkt. Min.)**
24. Ciszak L, Frydecka I, Wołowiec D, Szteblich A, Kosmaczewska A.: CTLA-4 affects expression of key cell cycle regulators of G0/G1 phase in neoplastic lymphocytes from patients with chronic lymphocytic leukaemia. *Clin Exp Med*, 2016, 16 (3): 317-332 **IF – 2,854 (25 pkt. Min.)**
25. Ciszak L, Frydecka I, Wołowiec D, Szteblich A, Kosmaczewska A.: Patients with chronic lymphocytic leukemia (CLL) differ in the pattern of CTLA-4 expression on CLL cells: the possible implications for immunotherapy with CTLA-4 blocking antibody. *Tumor Biol*, 2016, 37(3): 4143-4157 **IF - 2,926 (30 pkt. Min.)**
26. Dąbrowska K, Witkiewicz W. Correlations of host genetics and gut microbiome composition. *Frontiers in Microbiology*, 2016 vol. 7 art. 1357 **IF – 4,165 (35 pkt. Min.)**
27. Di Cesare M., Bentham J., et al. Koziel S., et al. Zuniga Cisneros J, (756 Collaborators): (NCD Risk Factor Collaboration). Trends in adult body-mass index in 200 countries from 1975 to 2014: a pooled analysis of 1698 population-based measurement studies with 19.2 million participants. *The Lancet,* 2016, 387: 1377-1396 **IF - 44,002 (50 pkt. Min.)**
28. Danel, D. P., Dziedzic-Danel, A., & Kleisner, K.: Does age difference really matter? Facial markers of biological quality and age difference between husband and wife. *HOMO – Journal of Comparative Human Biology*, 2016, 67(4), 337-347 **IF - 0,579 (15 pkt. Min.)**
29. Drab M, Krajniak J, Grzelakowski K.P: The new methodology and chemical kontrast observation by use of the energy-selective back-scattered electron detector. *Microscopy Microanalysis,* 2016, (6):1369-1373. doi: 10.1017/S1431927616012514 **IF – 1,730 (35 pkt. Min.)**
30. Drynda A, Obminska-Mrukowicz B, Kochanowska I, Zaczyńska E, Zimecki M, Mączyński M, Ryng S.: 5-amino-3-methyl-4-isoxazolecarboxylic acid hydrazide derivatives with in vitro immunomodulatory activities**.** *Chemical Biology & Drug Design*, 2016 Dec;68(12). doi: 10.1111/jphp.12631. **IF - 2,802 (25 pkt. Min.)**
31. Drynda A, Obmińska-Mrukowicz B, Zaczyńska E, Zimecki M, Mączyński Mn, Ryng S.: Immunoregulatory effects of 5-amino-3-methyl-4 isoxazolecarboxylic acid hydrazide and its derivative (4-(4-chlorophenyl)-1-(5-amino-3-methylisoxazole-4-carbonyl)-thiosemi-carbazide) in mice. *Journal of Pharmacy and Pharmacology*, 2016 Oct 26. doi: 10.1111/jphp.12631, 68(12):1613-1620 **F - 2,363 (25 pkt. Min.)**
32. Duan Z, Niedziela T, Lugowski C, Cao B, Wang T, Xu L, Yang B, Liu B, Wang L. Genetic Diversity of O-Antigens in Hafnia alvei and the Development of a Suspension Array for Serotype Detection. *PLoS One*, 2016; 11(5):e0155115. doi: 10.1371/journal. pone. 0155115. eCollection 2016. **IF - 3,057 (40 pkt. Min.)**

33. Dudek B., Krzyżewska E., Kapczyńska K., Rybka J., Pawlak A., Korzekwa K., Klausa E., Bugla-Płoskońska G. Proteomic analysis of outer membrane proteins from Salmonella Enteritidis strains with different sensitivity to human serum. *PLoS One* 2016 Oct. 3;11(10), doi: 10.1371/journal.pone.0164069 **IF – 3,057 (40 pkt. Min.)**
34. Eipper S, Steiner R, Lesner A, Sienczyk M, Palesch D, Halatsch ME, Zaczynska E, Heim Ch, Hartmann MD, Zimecki M, Wirtz ChR, Burster T.: Lactoferrin is an enhancer of the proteolytic activity of the innate defense protein Cathepsin G. *Plos One*, 2016, 11, doi.org/10.1371/journal.pone.0151509 **IF – 3,057 (40 pkt. Min.)**
35. Fidyt K., Fiedorowicz A., Strządała L., Szumny A.: β-caryophyllene and β-caryophyllene oxide-natural compounds of anticancer and analgesic properties. *Cancer Med*., 2016, 5: 3007-3017 **IF - 2,915 (25 pkt. Min.)**
36. Foerster A., Hołowacz I., SunilKumar G.B., Anandakumar S., Wall J.G., Wawrzynska M., Paprocka M., Kantor A., Kraskiewicz H., Olsztynska-Janus S., Hinder S.J., Bialy D., Podbielska H., Kopaczynska M.: Stainless steel surface functionalization for immobilization of antibody fragments for cardiovascular applications. *Biomed Mater Res A.*, 2016 Apr; 104(4):821-32. doi: 10.1002/jbm.a.35616. Epub 2015 Dec 15**. IF - 3,361 (30 pkt. Min.)**
37. Giese M, Turiello N, Molenda N, Palesch D, Meid A, Basilico P, Benarafa Ch, Halatsch M, Zimecki M, Westhoff MA, Wirtz ChR, Burster T.: Exogenous cathepsin G upregulates cell surface MHC class I molecules on immune and glioblastoma cells. *Oncotarget,* 2016, Oct 28. doi: 10.18632/oncotarget.12980. **IF - 5,008 (40 pkt. Min.)**
38. Gliszczyńska A, Gładkowski W, Świtalska M, Wietrzyk J, Szumny A, Gębarowska E, Wawrzeńczyk C.: Dehalogenation Activity of Selected Fungi Towards *δ*-Iodo-*γ*-Lactone Derived from *trans, trans*-Farnesol. *Chemistry & Biodiversity*, 2016; 13: 477–482. doi: 10.1002/cbdv.201500127 **IF – 1,444 (20 pkt. Min.)**
39. Gliszczyńska A, Niezgoda N, Gładkowski W, Czarnecka M, Świtalska M, Wietrzyk **J**.: Synthesis and Biological Evaluation of Novel Phosphatidylcholine Analogues Containing Monoterpene Acids as Potent Antiproliferative Agents. *PLoS One,* 2016, 11(6): e0157278. doi: 10.1371/journal.pone.0157278 **IF – 3,057 (40 pkt. Min.)**
40. Górska S, Buda B, Brzozowska E, Schwarzer M, Srutkova D, Kozakova H, Gamian A.: Identification of Lactobacillus proteins with different recognition patterns between immune rabbit sera and nonimmune mice or human sera. *BMC Microbiol,* 2016, 16:17. **IF – 2,581 (30 pkt. Min.)**
41. Górska S, Dylus E, Rudawska A, Brzozowska E, Srutkova D, Schwarzer M, Razim A, Kozakova H, Gamian A.: Immunoreactive Proteins of *Bifidobacterium longum* ssp. *longum* CCM 7952 and *Bifidobacterium longum* ssp. *longum* CCDM 372 Identified by Gnotobiotic Mono-Colonized Mice Sera, Immune Rabbit Sera and Non-immune Human Sera. *Front Microbiol*. 2016, 29;7:1537. **IF – 4,165 (35 pkt. Min.)**
42. Górska S, Hermanova P, Ciekot J, Schwarzer M, Srutkova D, Brzozowska E, Kozakova H, Gamian A.: Chemical characterization and immunomodulatory properties of polysaccharides isolated from probiotic Lactobacillus casei LOCK 0919. *Glycobiology,* 2016, 26(9):1014-1024. Erratum: 2016, 1–3 doi: 10.1093/glycob/cww107 **IF – 3,283 (30 pkt. Min.)**
43. Górska S, Sandstrőm C, Wojas-Turek J, Rossowska J, Pajtasz-Piasecka E, Brzozowska E, Gamian A.: Structural and immunomodulatory differences among lactobacilli exopolysaccharides isolated from intestines of mice with experimentally induced inflammatory bowel disease. *Scientific Reports*, 2016; 6: 37613 **IF – 5,228 (40 pkt. Min**.)
44. Górski A., Międzybrodzki R., Weber-Dąbrowska B., Fortuna W., Letkiewicz S., Rogóż P., Jończyk- Matysiak E., Dąbrowska K., Majewska J., Borysowski J.: Phage therapy: Combating infections with potential for evolving from merely a treatment for complications to targeting diseases. *Frontiers in Microbiology*, 2016, vol 7, art. 1515 **IF – 4,165 (35 pkt. Min.)**

45. Grzywa R, Winiarski Ł, Psurski M**,** Rudnicka A, Wietrzyk J, Gajda T, Oleksyszyn J.: Synthesis and biological activity of diisothiocyanate-derived mercapturic acids. *Bioorg Med Chem Lett*, 2016, 26(2):667-671. **IF – 2,486 (25 pkt. Min.)**
46. Hivelin M, Klimczak A, Cwykiel J, Sonmez E, Nasir S, Gatherwright J, Siemionow M.: Immunomodulatory Effects of Different Cellular Therapies of Bone Marrow Origin on Chimerism Induction and Maintenance Across MHC Barriers in a Face Allotransplantation Model. *Arch Immunol Ther Exp*, 2016 Aug; 64(4):299-310. doi: 10.1007/s00005-015-0380-8. Epub 2015 Dec 26. **IF - 2,464 (25 pkt. Min.)**
47. Huczyński A, Majcher U, Maj E, Wietrzyk J, Janczak J, Moshari M, Tuszynski JA, Bartl F.: Synthesis, antiproliferative activity and molecular docking of Colchicine derivatives *Bioorganic Chemistry*, 2016, 64: 103-112. **IF – 2,252 (20 pkt. Min.)**
48. Ignasiak, Z., Falkenberg, J., Ignasiak T., Koziel, S.: The influence of a 7-month-long intensive aquatic exercise program on changes in physical fitness, bone mineral density and lung function parameters in postmenopausal women. *Collegium Antropologicum*, 2016, 40(4): **IF - 0,609 (25 pkt. Min.)**
49. Ipsen, J., Nowak-Szczepanska, N., Gomula, A., Aßmann, C., Hermanussen, M.: The association of body height, height variability and inequality. *Anthropologischer Anzeiger*, 2016, 73(1):1-6 **IF – 0,426 (15 pkt. Min.)**
50. Iwaszko M., Świerkot J., Kolossa K., Jeka S., Wiland P., Bogunia-Kubik K.: Influence of CD94 and NKG2A variants on susceptibility to rheumatoid arthritis and efficacy of anti-TNF treatment. *Joint Bone Spine*, 2016; 83(1): 75-9. **IF – 2,946 (25 pkt. Min.)**
51. Jasek M, Bojarska-Junak A, Wagner M, Sobczyński M, Wołowiec D, Roliński J, Karabon L, Kuśnierczyk P. Association of variants in BAFF (rs9514828 and rs1041569) and BAFF-R (rs61756766) genes with the risk of chronic lymphocytic leukemia.*Tumor Biol*, 2016;37: 13617–13626. DOI 10.1007/s13277-016-5182-z. PMID: 27468724 **IF – 2,926 (30 pkt. Min.)**
52. Jaworski P., Donczew R., Mielke T., Thiel M., Oldziej S., Weigel C., Zawilak-Pawlik A.: Unique and universal features of Epsilonproteobacterial origins of chromosome replication and DnaA-DnaA box interactions. *Front. Microbiol*, 2016, 7:1555. doi: 10.3389/fmicb.2016.01555. **IF - 4,165 (35 pkt. Min.)**
53. Jeleń M, Pluta K, Zimecki M, Morak–Młodawska B, Artym J, Kocięba M, Kochanowska I.: Synthesis and biological evaluation of novel propargylquinobenzothiazines and their derivatives as potential antiproliferative, anti-inflammatory and anticancer agents**.** *Journal of Enzyme Inhibition and Medicinal Chemistry*, 2016, 12, 1-6; .doi.org/10.1080 /14756366.2016.1205046 **IF – 3,428 (20 pkt. Min.)**
54. Kaczmarek R, Duk M, Szymczak K, Korchagina E, Tyborowska J, Mikolajczyk K, Bovin N, Szewczyk B, Jaskiewicz E, Czerwinski M.: Human Gb3/CD77 synthase reveals specificity toward two or four different acceptors depending on amino acid at position 211, creating P-k, P1 and NOR blood group antigens. *Biochem Biophys Res Commun*, 2016, 470, (1), 168-74 **IF – 2,371 (20 pkt. Min.)**
55. Kaczmarek R, Mikolajewicz K, Szymczak K, Duk M, Majorczyk E, Krop-Watorek A, Buczkowska A, Czerwinski M.: Evaluation of an amino acid residue critical for the specificity and activity of human Gb3/CD77 synthase. *Glycoconj J.* 2016, 33(6), 963-973 **IF - 1,828 (20 pkt. Min.)**
56. Kalaska B, Kamiński K, Mikosz J, Yusa S, Sokolowska E, Błażejczyk A, Wietrzyk J, Lewandowska A, Kasacka I, Szczubialka K, Pawlak D, Nowakowska M, Mogielnicki A.: Heparin-binding copolymer (HBC) reverses effects of unfractionated heparin, enoxaparin and fondaparinux in rats and mice. *Translational Research***,** 2016, 177:98-112.e10. **IF – 4,557 (40 pkt. Min.)**

57. Kałuża A, Jarząb A, Gamian A, Kratz EM, Zimmer M, Ferens-Sieczkowska M.: Preliminary MALDI-TOF-MS analysis of seminal plasma N-glycome of infertile men. *Carbohydr Res*, 2016, 435:19-25. **IF – 1,817 (25 pkt. Min.)**
58. Kamińska K, Wojaczyńska E, Wietrzyk J, Turlej E, Błażejczyk A**,** Wieczorek R.: Synthesis, structure and antiproliferative activity of chiral polyamines based on a 2-azanorbornane skeleton. *Tetrahedron: Asymmetry*, 2016, 27 (16), 753–758. **IF – 2,108 (25 pkt. Min.)**
59. Karabon L, Partyka A, Jasek M, Lech–Maranda E, Grzybowska–Izydorczyk O, Bojarska–Junak A, Pawlak–Adamska E, Tomkiewicz A, Robak T, Rolinski J and Frydecka I: Intragenic variations in BTLA gene influence mRNA expression of BTLA gene in chronic lymphocytic leukaemia patients and confer susceptibility to chronic lymphocytic leukaemia. *Arch. Immunol. Ther. Exp.*, 2016, 64, suppl. 1, 137-146 **IF – 2,464 (25 pkt. Min.)**
60. Kaszowska M, Stojkovic K, Niedziela T, Lugowski C.: The O-antigen of Plesiomonas shigelloides serotype O36 containing pseudaminic acid. *Carbohydr Res.*, 2016; 434:1-5. doi: 10.1016/j.carres.2016.07.007. **IF - 1,817 (25 pkt. Min.)**
61. Kasztura M., Sniezewski L., Laszkiewicz A., Majkowski M., Kobak K., Peczek K., Janik S., Kapusniak V., Miazek A., Cebrat M., Kisielow P.: Search for the function of NWC, third gene within RAG locus: generation and characterization of NWC deficient mice. *Arch Immunol Ther Exp*, 2016, 64, 311–319 **IF - 2,464 (25 pkt. Min.)**
62. Kicielińska J, Szczygieł A, Rossowska J, Anger N, Kempińska K, Świtalska M, Kaszowska M, Wietrzyk J, Boratyński J, Pajtasz-Piasecka E.: Oral Administration of Polymyxin B Modulates the Activity of Lipooligosaccharide E. coli B against Lung Metastases in Murine Tumor Models. *PLoS One* **2016**, 11(2): e0148156. doi:10.1371/journal.pone.0148156 **IF – 3,057 (40 pkt. Min.)**
63. Klekotko M., Matczyszyn K., Siednienko J.,Olesiak-Bańska J., Pawlik K., Samoć M. Environmentally friendly synthesis of gold nanoparticles. 2016, *SPIE Newsroom*. DOI: 10.1117/2.1201512.006210 IF - 0 (0 pkt. Min.)
64. Klimczak A, Kozłowska U.: Mesenchymal Stromal Cells and Tissue-Specific Progenitor Cells: Their Role in Tissue Homeostasis. *Stem Cells Int.,* 2016; 4285215. doi: 10.1155/2016/4285215. Epub 2015 Dec 28*;* **IF - 3,687 (20 pkt. Min.)**
65. Koba M, Śmietana M, Brzozowska E, Górska S, Janik M, Mikulic P, Cusano A, Bock W.J.. Bacteriophage Adhesin-Coated Long-Period Grating-Based Sensor: Bacteria Detection Specificity. *Journal of Lightwave Technology*, 2016, 34(19), 4531-4536 DOI: 10.1109/ JLT.2016.2532466. **IF - 2.567 (35 pkt. Min.)**
66. Kociuba M, Koziel S, Chakraborty R.: Sex differences in digit ratio (2D:4D) among the students of military and civil courses at military academy in Wroclaw, Poland. *Journal of Biosocial Science,* 2016, 48(5): 658-671 **IF – 1,056 (20 pkt. Min.)**
67. Koziel, S., Kociuba, M., Ignasiak, Z., Chakraborty, R.: Is sport choice and participation related to 2D:4D? A study among adult male student in Wroclaw, Poland. *Collegium Antropologicum,* 2016, 40(2): 105-110 **IF – 0,609 (25 pkt. Min.)**
68. Król J, Wanecka A, Twardoń J, Mrowiec J, Dropińska A, Bania J, Podkowik M, Korzeniowska-Kowal A, Paściak M. Isolation of Staphylococcus microti from milk of dairy cows with mastitis *Vet. Microbiol*, 2016, 182, 163-169 **IF – 2,564 (35 pkt. Min.)**
69. Krüger NJ., Knüve MT., Zawilak-Pawlik A., Appel B., Stingl K. Genetic Diversity as Consequence of a microaerobic and neutrophilic lifestyle., *PLOS Pathog,* 2016, 12: e1005626. doi:10.1371/journal.ppat.1005626 **IF - 7,003 (45 pkt. Min.)**
70. Krzystek-Korpacka M, Gorska S, Diakowska D, Kapturkiewicz B, Podkowik M, Gamian A, Bednarz-Misa I.: Midkine is up-regulated in both cancerous and inflamed bowel, reflecting lymph node metastasis in colorectal cancer and clinical activity of ulcerative colitis. *Cytokine,* 2016, Sep 28. 89, 68-75 **IF – 2,940 (25 pkt. Min.)**

71. Książczyk M, Kuczkowski M, Dudek B, Korzekwa K, Tobiasz A, Korzeniowska-Kowal A, Paluch E, Wieliczko A, Bugla-Płoskońska G.: Application of Routine Diagnostic Procedure, VITEK 2 Compact, MALDI-TOF MS, and PCR Assays in Identification Procedure of Bacterial Strain with Ambiguous Phenotype. *Curr Microbiol.* 2016 May;72(5):570-82. doi: 10.1007/s00284-016-0993-0. **IF – 1,519 (15 pkt. Min.)**
72. Kuebler WM, Wittenberg C, Lee WL, Reppien E, Goldenberg NM, Lindner K, Gao Y, Winoto-Morbach S, Drab M, Mühlfeld C, Dombrowsky H, Ochs M, Schütze S, Uhlig S. Thrombin stimulates albumin transcytosis in lung microvascular endothelial cells via activation of acid sphingomyelinase. *Am J Physiol Lung Cell Mol Physiol*. 2016 Apr 15;310(8):L720-32. doi: 10.1152/ajplung.00157.2015. PMID: 26851257, **IF - 4.721 (40 pkt. Min.)**
73. Kulbacka J, Pucek A, Kotulska M, Dubińska-Magiera M, Rossowska J, Rols MP, Wilk KA.: Electroporation and lipid nanoparticles with cyanine IR-780 and flavonoids as efficient vectors to enhanced drug delivery in colon cancer. *Bioelectrochemistry*. 2016, 110:19-31, **IF - 3,556 (35 pkt. Min.)**
74. Kulbacka J, Pucek A, Wilk KA, Dubińska-Magiera M, Rossowska J, Kulbacki M, Kotulska M.: The Effect of Millisecond Pulsed Electric Fields (msPEF) on Intracellular Drug Transport with Negatively Charged Large Nanocarriers Made of Solid Lipid Nanoparticles (SLN): In Vitro Study. *J Membr Biol*., 2016 Oct; 249(5):645-661. **IF - 1,991 (20 pkt. Min.)**
75. Kupczyk P, Reich A, Hołysz M, Gajda M, Wysokińska E, Kobuszewska A, Nevozhay D, Nowakowska B, Strzadała L, Jagodziński P, Szepietowski J,C.: Opioid Receptors in Psoriatic Skin: Relationship with Itch. *Acta Derm Venereol*. 2016 Dec 13. doi: 10.2340/00015555 **IF – 3,638 (40 pkt.)**
76. Lamch Ł, Kulbacka J, Pietkiewicz J, Rossowska J, Dubińska-Magiera M, Choromańska A, Wilk KA.: Preparation and characterization of new zinc(II) phthalocyanine - Containing poly(l-lactide)-b-poly(ethylene glycol) copolymer micelles for photodynamic therapy. *J Photochem Photobiol B*. 2016 Jul; 160:185-97. **IF - 3,035 (30 pkt. Min.)**
77. Lechowicz L, Chrapek M, Czerwonka G, Korzeniowska-Kowal A, Tobiasz A, Urbaniak M, Matuska-Lyzwa J, Kaca W.: Detection of ureolytic activity of bacterial strains isolated from entomopathogenic nematodes using infrared spectroscopy. *J Basic Microbiol*. 2016;56(8):922-8. doi: 10.1002/jobm.201500538. **IF – 1,585 (15 pkt. Min.)**
78. Lemieszewska M, Jakubik-Witkowska M, Stańczykiewicz B, Bambrowicz A, Zabłocka A, Polanowski A, Trziszka T, Rymaszewska J: Pro-Cognitive properties of the immunomodulatory polypeptide complex, Yolkin, from chicken egg yolk and colostrum-derived substances: analyses based on animal model of age-related cognitive deficits. *Arch Immunol Ther Exp*, 2016, 64, 425-434; DOI 10.1007/s00005-016-0392-z **IF – 2,464 (25 pkt. Min.)**
79. Lipowicz A., Szklarska A., Mitas W.A.: Biological costs of economic transition: stress levels during the transition from Communism to Capitalism in Poland. *Economics and Human Biology*, 2016, 21: 90-99 **IF – 1,639 (35 pkt. Min.)**
80. Lisowska-Gaczorek A., Koziel S., Cienkosz-Stepańczak B., Mądrzyk K., Pawlyta S., Gronkiewicz S., Wołoszyn M., Szostek K.: An analysis of the origin of an early medieval group of individuals from Gródek based on the analysis of stable oxygen isotopes. *HOMO – Journal of Comparative Human Biology*, 2016, 67(4): 313-327 **IF – 0,579 (25 pkt. Min.)**
81. Liu, T.-M., Conde, J., Lipiński, T., Bednarkiewicz, A., & Huang, C.-C. Revisiting the classification of NIR-absorbing/emitting nanomaterials for *in vivo* bioapplications. 2016, NPG Asia Materials, 8(8), e295. **IF - 8,772 (45 pkt. Min.)**
82. Łakomska I, Stefańczak K, Fandzloch M, Sitkowski J, Filip-Psurska B, Wojtczak A.: Use of 1,2,4-triazolo[1,5-a]pyrimidines to design new "piano-stool" ruthenium(II) compounds, *Polyhedron*, 2016, 109: 33-39. **IF – 2,108 (25 pkt. Min.)**

83. Łączkowski KZ, Świtalska M, Baranowska-Łączkowska A, Plech T, Paneth A, Misiura K, Wietrzyk J, Czaplińska B, Mrozek-Wilczkiewicz A, Malarz K, Musioł R, Grela I.: Thiazole-based nitrogen mustards: Design, synthesis, spectroscopic studies, DFT calculation, molecular docking, and antiproliferative activity against selected human cancer cell lines. *J Mol Struct*, 2016, 1119: 139-150 **IF – 1,708 (20 pkt. Min.)**
84. Łukasik E, Waśniowska K.: Antygeny układu Duffy: budowa, właściwości serologiczne i funkcja. *Post Hig Med Dośw,* 2016, 70, 143-161 **IF – 0,769 (15 pkt. Min.)**
85. Mączyński M, Artym J, Kocięba M, Kochanowska I, Ryng S, Zimecki M.: Anti-inflammatory properties of an isoxazole derivative - MZO-2 compound. *Pharmacol Reports*, 2016, 68, 894-902; doi.org/10.1016/j.pharep.2016.04.017 **IF – 2,251 (25 pkt. Min.)**
86. Mączyński M, Artym J, Kocieba M, Sochacka-Ćwikla A, Drozd-Szczygieł E, Ryng S, Zimecki M.: Synthesis and Immunoregulatory properties of selected 5-amino-3-methyl 4-isoxazolecarboxylic acid benzylamides. *Acta Poloniae Pharmaceutica,* 2016, 73, 1201-1211; **IF - 0,877 (15 pkt. Min.)**
87. Maj E, Papiernik D, Wietrzyk J.: Antiangiogenic Cancer Treatment – the Great Discovery and Greater Complexity. *Int J Oncol*., 2016, 49: 1773-1784. **IF – 3,018 (25 pkt. Min.)**
88. Makowski Ł., Donczew R., Weigel C., Zawilak-Pawlik A., Zakrzewska-Czerwinska J.: Initiation of chromosomal replication in predatory bacterium *Bdellovibriobacteriovorus*. *Front Microbiol*, 2016, 7:1898. doi: 10.3389/fmicb.2016.01898. **IF - 4,165 (35 pkt. Min.)**
89. Man-Kupisinska A, Bobko E, Gozdziewicz TK, Maciejewska A, Jachymek W, Lugowski C, Lukasiewicz J.: Fractionation and analysis of lipopolysaccharide-derived oligosaccharides by zwitterionic-type hydrophilic interaction liquid chromatography coupled with electrospray ionisation mass spectrometry. *Carbohydr Res*, 2016; 427:29-37. doi: 10.1016/j.carres. 2016.03.024. **IF - 1,817 (25 pkt. Min.)**
90. Man-Kupisinska A, Michalski M, Maciejewska A, Swierzko AS, Cedzynski M, Lugowski C, Lukasiewicz J.: A New Ligand-Based Method for Purifying Active Human Plasma-Derived Ficolin-3 Complexes Supports the Phenomenon of Crosstalk between Pattern-Recognition Molecules and Immunoglobulins. *PLoS One*, 2016; 11(5):e0156691. doi:10.1371/journal.pone. 0156691. eCollection, 2016 **IF - 3,057 (40 pkt. Min.)**
91. Marchwicka A., Cebrat M., Laszkiewicz A., Sniezewski L., Brown G., Marcinkowska E.: Regulation of vitamin D receptor expression by retinoic acid receptor alpha in acute myeloid leukemia cells. *J Steroid Biochem Mol Biol*, 2016, 159, 121-130 **IF - 3,985 (30 pkt. Min.)**
92. Marek-Bukowiec K., Aguado E., Miazek A.: Phorbol ester-mediated re-expression of endogenous LAT adapter in J.CaM2 cells: a model for dissecting drivers and blockers of LAT transcription. *Genes and Immunity*, 2016, 17, 313-320 **IF - 2,472 (30 pkt. Min)**
93. Matysiak J, Skrzypek A, Karpińska M, Głaszcz U, Niewiadomy A, Paw B, Sęczyna B, Wietrzyk J, Kłopotowski D.: Synthesis and antiproliferative activity of some N′-substituted 2,4-dihydroxybenzothiohydrazides. *J Enzyme Inhibition and Medicinal Chemistry*. 2016, doi: 10.1080/14756366.2016.1212192 **IF – 3,428 (20 pkt. Min.)**
94. Matysiak J, Skrzypek A, Niewiadomy A, Karpińska M.M, Wietrzyk J, Paw B, Kłopotowska D.: Synthesis of 4-(4-Methylidene-4*H*-3,1-Benzothiazin-2-yl)benzene-1,3-Diols and Their Antiproliferative Activity Against Human Cancer Cell Lines. *Russ J Bioorg Chem*, 2016, 42 (1): 104-110 **IF – 0,660 (15 pkt. Min.)**
95. Mieczkowski A, Tomczyk E, Makowska MA, Nasulewicz-Goldeman A, Gajda R, Woźniak K, Wietrzyk J.: Synthesis and Investigation of the Antitumor Properties of Novel, Bicyclic Furopyrimidine, Pyrrolopyrimidine and Pyrimidopyridazine Nucleoside Analogues, *Synthesis*, 2016, 48, 566-572 **IF – 2,652 (30 pkt. Min.)**

96. Miernikiewicz P, Kłopot A, Soluch R, Szkuta P, Kęska W, Hodyra-Stefaniak K, Konopka A, Nowak M, Lecion D, Kaźmierczak Z, Majewska J, Harhala M, Górski A, Dąbrowska K. T4 phage tail adhesin gp12 counteracts LPS-induced inflammation in vivo. Frontiers in Microbiology 2016, vol. 7:1112 **IF – 4,165 (35 pkt. Min.)**
97. Mikołajczyk K., Kaczmarek R., Czerwiński M.: Czy mutacje w genie kodującym czynnik transkrypcyjny EKLF (Erythroid Krüppel-Like Factor) mogą chronić nas przed chorobami zakaźnymi i pasożytniczymi? *Post Hig Med Dośw,* 2016, 70, 1068-1086 **IF – 0,769 (15 pkt. Min.)**
98. Mozer-Lisewska I, Zwolińska K, Kowala-Piaskowska A.E, Bura M, Rozpłochowski B, Pauli A, Żeromski J, Piasecki E, Kuśnierczyk P.: Genetic (KIR, HLA-C) and some clinical parameters influencing the level of liver enzymes and early virologic response in patients with chronic hepatitis C. *Arch. Immunol. Ther. Exp.*, 2016, 64: 65-73 **IF – 2,464 (25 pkt. Min.)**
99. Myga-Nowak M, Godela A, Głąb T, Lewańska M, Boratyński J.: Bacteriophages to combat foodborne infections caused by food contamination by bacteria of the Campylobacter genus. 2016, *Post Hig Med Dosw,* (online), 2016, 70: 989-1000. doi:10.5604/17322693.1220084 **IF – 0,769 (15 pkt.)**
100. Nowak I, Malinowski A, Barcz E, Wilczyński JR, Wagner M, Majorczyk E, Motak-Pochrzęst H, Banasik M, Kuśnierczyk P.: Possible role of HLA-G, LILRB1 and KIR2DL4 gene polymorphisms in spontaneous miscarriage. *Arch Immunol Ther Exp*, 2016, 64(6): 505-514 doi: 10.1007/s00005-016-0389-7 **IF - 2,464 (25 pkt. Min.)**
101. Nowak-Szczepanska N., Koziel S.: Sexual dimorphism in growth in the relative length of the forearm and relative knee height during adolescence. *American Journal of Physical Anthropology*, 2016, 161, (2): 276-282 **IF – 2,402 (30 pkt. Min.)**
102. Nowak-Szczepanska, N., Gomula, A., Ipsen, M. J., Koziel, S.: Different effects of living conditions on the variation in BMI and height in children before the onset of puberty. *European Journal of Clinical Nutrition*, 2016, 70(6):662-666 **IF – 2,935 (30 pkt. Min.)**
103. Pacia M.Z, Mateuszuk Ł, Buczek E, Chłopicki S, Błażejczyk A, Wietrzyk J, Baranska M, Kaczor A.: Rapid biochemical profiling of endothelial dysfunction in diabetes, hypertension and cancer metastasis by hierarchical cluster analysis of Raman spectra. *J. Raman Spectrosc*., 2016, 47(11), 1310-1317 doi:10.1002/jrs.4965. **IF – 2,395 (30 pkt. Min.)**
104. Pacia MZ, Mateuszu Ł, Buczek E, Chłopicki S, Blazejczyk A, Wietrzyk J, Baranska M, Kaczor A.: 3D Raman imaging of systemic endothelial dysfunction in the murine model of metastatic breast cancer. *Analytical and Bioanalytical Chemistry*, 2016, 408(13), 3381-3387, doi: 10.1007/s00216-016-9436-9 **IF – 3,125 (35 pkt. Min.)**
105. Palesch D, Wagner J, Meid A, Molenda N, Sienczyk M, Burkhardt J, Münch J, Prokop L, Stevanovic S, Westhoff MA, Halatsch ME, Wirtz Ch.R, Zimecki M, Burster T.: Cathepsin G-mediated proteolytic degradation of MHC class I molecules to facilitate immune detection of human glioblastoma cells. *Cancer Immunol Immunothe,* 2016; 65, 283-291**.** doi: 10.1007/s00262-016-1798-5. Epub 2016 Feb 2; **IF – 4,846 (30 pkt. Min.)**
106. Partyka A, Tupikowski K, Kolodziej A, Zdrojowy R, Halon A, Malkiewicz B, Dembowski J, Frydecka I, Karabon L.: Association of 3' nearby gene BTLA polymorphisms with the risk of renal cell carcinoma in the Polish population. *Urol Oncol*, 2016, 34(9): 419.e13-9. **IF – 2,921 (30 pkt. Min.)**
107. Paściak M, Górska S, Jawiarczyk N, Gamian A.: Lactobacillus johnsonii glycolipids, their structure and immunoreactivity with sera from inflammatory bowel disease patients. *Microb. Biotechnol*, 2016, 2016 Oct 21. doi: 10.1111/1751-7915.12424. **IF – 3,991 (35 pkt. Min.)**
108. Pawlak A., Obmińska-Mrukowicz B., Zbyryt I., Rapak A.: In vitro drug sensitivity in canine lymphoma. *J. Vet. Res*, (Bull Vet. Inst. in Pulawy) 2016, 60: 55-61 **IF – 0,300 (15 pkt. Min.)**

109. Pawlak A, Rapak A, Drynda A, Paradowski D, Zbyryt I, Dzimira S, Suchański J, Obmińska-Mrukowicz B.: Immunophenotypic characterization of canine malignant lymphoma: a retrospective study of cases diagnosed in Poland Lower Silesia, over the period 2011–2013. *Vet Com Oncol*, 2016, 14(1), 52-60 **doi: 10.1111/vco.12112 IF – 2,452 (45 pkt.)**
110. Pawlak A., Zioło E., Fiedorowicz A., Fidyt K., Strzadała L., Kałas W., Long-lasting reduction in clonogenic potential of colorectal cancer cells by sequential treatments with 5-azanucleosides and topoisomerase inhibitors. *BMC Cancer*, 2016, 16(1) 893 (13 stron) **IF - 3,265 (30 pkt. Min.)**
111. Pawlak A., Zioło E., Kutkowska J., Błażejczyk A., Wietrzyk J., Krupa A., Hildebrand W., Dzięgiel P., Dzimira S., Obmińska-Mrukowicz B., Strządała L., Rapak A.: A novel canine B-cell leukaemia cell line. Establishment, characterisation and sensitivity to chemotherapeutics. *Vet. Comp. Oncol*., 2016, Aug 9. doi: 10.1111/vco.12257. **IF - 2,452 (45 pkt. Min.)**
112. Podbielska M, Das A, Smith AW, Chauhan A, Ray SK, Inoue J, Azuma M, Nozaki K, Hogan EL and Banik NL.: Neuron-microglia interaction induced bi-directional cytotoxicity associated with calpain activation. *J Neurochem,* 2016, 139(3): 440-455 **IF – 3,842 (30 pkt. Min.)**
113. Podbielska M, Szulc ZM, Kurowska E, Hogan EL, Bielawski J, Bielawska A and Bhat NR.: Cytokine-induced release of ceramide-enriched exosomes as a mediator of cell death signaling in an oligodendroglioma cell line. *J Lipid Res*, 2016, 57(11): 2028-2039 **IF – 4,368 (35 pkt. Min.)**
114. Przygodzki T, Talar M, Błażejczyk A, Kalchenko V, Watala C.: Quantification of the Blood Platelet Reactivity in the ADP-Induced Model of Non-Lethal Pulmonary Thromboembolism in Mice with the Use of Laser Doppler Flowmetry. *PLoS One,* 2016; 11(1):e0146346. doi: 10.1371/journal.pone.0146346. **IF – 3,057 (40 pkt. Min.)**
115. Reniewicz P, Zyzak J, Siednienko J.: Komórkowe receptory egzogennego RNA. *Post Hig Med. Dośw,* (online), 2016, 70, 337-48 **IF – 0,769 (15 pkt. Min.)**
116. Rogala P, Jabłońska-Wawrzycka A, Kazimierczuk K, Borek A, Błażejczyk A, Wietrzyk J, Barszcz B.: Synthesis, crystal structure and cytotoxic activity of ruthenium(II) piano-stool complex with N,N-chelating ligand. *J Mol Struct.* 2016, 1126: 74-82. **IF – 1,708 (20 pkt. Min.)**
117. Rogatko-Koros M., Mika-Witkowska R., Bogunia-Kubik K., Wysoczanska B., Jaskuła E., Koscinska K., Nestorowicz K., Dziopa J., Szlendak U., Gwozdowicz S., Graczyk-Pol E., Lange A. Nowak J.: Prediction of NK cell licensing level in selection of hematopoietic stem cell donor - initial results. *Arch Immunol Ther Exp,* 2016, 64, 63-72 suppl. 1 **IF - 2,464 (25 pkt. Min.)**
118. Rybka J., Gębura K., Wróbel T., Wysoczańska B., Stefanko E., Kuliczkowski K., Bogunia-Kubik K.: Variations in genes involved in regulation of the nuclear factor - κB pathway and the risk of acute myeloid leukaemia. *Int J Immunogenet*, 2016; 43(2): 101-6. IF - 1,174 **IF – 1,174 (15 pkt. Min.)**
119. Rydosz A, Brzozowska E, Górska S, Wincza K, Gamian A, Gruszczynski S.: A broadband capacitive sensing method for label-free bacterial LPS detection. *Biosens Bioelectron,* 2016, 75:328-36. **IF – 7,476 (40 pkt. Min.)**
120. Skowicki M, Lipiński T.: The development of methods for obtaining monoclonal antibody-producing cells. *Post Hig Med Dosw*, (online), 2016, Apr 27;70:367-79 **IF – 0,769 (15 pkt. Min.)**
121. Sokolowska, Kalaska B, Kamiński K, Lewandowska A, Błażejczyk A, Wietrzyk J, Kasacka I, Szczubialka K, Pawlak D, Nowakowska M, Mogielnicki A.:. The toxicokinetic profile of Dex40-GTMAC3 - a novel polysaccharide candidate for reversal of unfractionated heparin. *Frontiers in Pharmacology*, 2016, 7:60. doi: 10.3389/fphar. 2016.00060 **IF – 4,418 (35 pkt. Min.)**

122. Sozański T., Kucharska A.Z., Rapak A., Szumny D., Trocha M., Merwid-Ląd A., Dzimira S., Piasecki T., Piórecki N., Magdalan J., Szeląg A.: Iridoid-loganic acid versus anthocyanins from the Cornus mas fruits (cornelian cherry): Common and different effects on diet-induced atherosclerosis, PPARs expression and inflammation. *Atherosclerosis*, 2016, 254: 151-160. doi:10.1016/j.atherosclerosis.2016.10.00 **IF - 3,942 (35 pkt. Min.)**

123. Spinek A.E., Lorkiewicz W., Mietlińska J., Sewerynek E., Kłys A., Caramelli D., Żądzińska E., Evaluation of chronological changes in bone fractures and age-related bone loss: A test case from Poland. *Journal of Archaeological Science*, 2016, 72, 117-127 **IF – 2,255 (35 pkt. Min.)**

124. Spólnicka M, Piekarska RZ, Jaskuła E, Basak GW, Jacewicz R, Pięta A, Makowska Ż, Jedrzejczyk M, Wierzbowska A, Pluta A, Robak T, Berent J, Branicki W,Jędrzejczak W, Lange A, Płoski R. Donor age and C1orf132/MIR29B2C determineage-related methylation signature of blood after allogeneic hematopoietic stemcell transplantation. *Clin Epigenetics.* 2016 Sep 6; 8:93. doi:10.1186/s13148-016-0257-7. Erratum in: *Clin Epigenetics.* 2016 Nov 18; 8:121. **IF - 4,327 (30 pkt. Min.)**

125. Stosio M, Witkowicz A, Kowalska A, **Karabon L**. Genetic background of aberrant thermogenin expression (UCP1) in obesity leading to metabolic syndrome. *Post Hig Med Dosw* (Online). 2016 Dec 31; 70(0):1389-1403. doi: 10.5604/17322693.1227677. **IF - 0,769 (15 pkt. Min.)**

126. Świerkot J, Nowak B, Czarny A, Zaczyńska E, Sokolik R, Madej M, Korman L, Sebastian A, Wojtala P, Lubiński Ł, Wiland P.: The Activity of JAK/STAT and NF-κB in Patients with Rheumatoid Arthritis. *Adv Clin Exp Med*, 2016, 25, 709-717; doi: 10.17219/acem/61034 **IF – 1,127 (15 pkt. Min.)**

127. Świerzko AS, Bartłomiejczyk MA, Brzostek A, Łukasiewicz J, Michalski M, Dziadek J, Cedzyński M. Mycobacterial antigen 85 complex (Ag85) as a target for ficolins and mannose-binding lectin. *Int J Med Microbiol*, 2016; 306(4):212-21. doi: 10.1016/j.ijmm.2016.04.004. **IF - 3,898 (35 pkt. Min.)**

128. Szczeszak A, Ekner-Grzyb A, Runowski M, Szutkowski K, Mrówczyńska L, Kaźmierczak Z, Grzyb T, Dąbrowska K, Giersig M, Lis S. Spectroscopic, structural and in vitro cytotoxicity evaluation of luminescent, lanthanide doped core@shell nanomaterials GdVO4:Eu(3+)5%@SiO2@NH2. *J Colloid Interf Sci*, 2016, 481: 245-55 **IF – 3,782 (30 pkt. Min.)**

129. Szijártó V, Guachalla LM, Hartl K, Varga C, Banerjee P, Stojkovic K, Kaszowska M, Nagy E, Lukasiewicz J, Nagy G. Both clades of the epidemic KPC-producing Klebsiella pneumoniae clone ST258 share a modified galactan O-antigen type. *Int J Med Microbiol*, 2016; 306(2):89-98. doi: 10.1016/j.ijmm.2015.12.002. **IF - 3,898 (35 pkt. Min.)**

130. Ugorski M., Dziegiel P., Suchanski J.: Podoplanin – a small glycoprotein with many faces. *Am. J. Cancer Res*, 2016, 6: 370-386 **IF – 3,425 (35 pkt. Min.)**

131. Weber-Dąbrowska B., Jończyk-Matysiak E., Żaczek M., Łobocka M., Łusiak-Szelachowska M., Górski A.: Bacteriophage procurement for therapeutic purposes. *Frontiers in Microbiology*, 2016, vol 7, art. 1177 **IF – 4,165 (35 pkt. Min.)**

132. Wiśniewski J, Wesołowska O, Środa-Pomianek K, Paprocka M, Bielawska-Pohl A, Krawczenko A, Duarte N, Ferreira MJ, Duś D, Michalak K.: Euphorbia Species-derived Diterpenes and Coumarins as Multidrug Resistance Modulators in Human Colon Carcinoma Cells. *Anticancer Res*., 2016 May; 36(5):2259-64. **IF – 1,895 (20 pkt. Min.)**

133. Wittmann A, Lamprinaki D, Bowles KM, Katzenellenbogen E, Knirel YA, Whitfield C, Nishimura T, Matsumoto N, Yamamoto K, Iwakura Y, Saijo S, Kawasaki N.: Dectin-2 Recognizes Mannosylated O-antigens of Human Opportunistic Pathogens and Augments Lipopolysaccharide Activation of Myeloid Cells. *J Biol Chem*, 2016; 291(34):17629-38. doi: 10.1074/jbc.M116.741256. **IF - 4,258 (35 pkt. Min.)**

134. Wojas-Turek J, Szczygieł A, Kicielińska J, Rossowska J, Piasecki E, Pajtasz-Piasecka E.: Treatment with cyclophosphamide supported by various dendritic cell-based vaccines induces diversification in CD4+ T cell response against MC38 colon carcinoma. *Int J Oncol*, 2016; 48(2):493-505 **IF – 3,018 (25 pkt. Min.)**

135. Wojcieszak D, Mazur M, Kaczmarek D, Mazur P, Szponar B, Domaradzki J, Kepinski L.: Influence of the surface properties on bactericidal and fungicidal activity of magnetron sputtered Ti-Ag and Nb-Ag thin films. *Mater Sci Eng C Mater Biol Appl*, 2016 62:86-95. doi: 10.1016/j.msec.2016.01.029. **IF – 3,420 (30 pkt. Min.)**

136. Wojcieszak D., Mazur M., Kaczmarek D., Poniedziałek A., Domanowski P., Szponar B., Czajkowska A., Gamian A. Effect of the structure on biological and photocatalytic activity of transparent titania thin-film coatings. *Materials Science-Poland*, 2016, 34(4), 856-862 DOI: 10.1515/msp-2016-0100. **IF – 0,536 (15 pkt. Min.)**

137. Wojcieszak D, Mazur M, Kaczmarek D, Szponar B, Grobelny M, Kalisz M, Pelczarska A, Szczygieł I, Poniedziałek A, Osekowska M.: Structural and surface properties of semitransparent and antibacterial (Cu,Ti,Nb)Ox coating. *Applied Surface Science*, 2016, 380: 159-164 DOI: 10.1016/j.apsusc.2016.01.232 **IF – 3,150 (35 pkt. Min.)**

138. Wysokińska E., Cichos J., Zioło E., Bednarkiewicz A., Strządała L., Karbowiak M., Hreniak D., Kałas W.: Cytotoxic interactions of bare and coated NaGdF4:Yb3+:Er3+ nanoparticles with macrophage and fibroblast cells. *Toxicology in Vitro*, 2016, 32, 16-25 **IF - 3,338 (30 pkt. Min.)**

139. Yao Y, Wiśniewski A, Ma Q, Kowal A, Porębska I, Pawełczyk K, Yu J, Dubis J, Żuk N, Li Y, Shi L, Kuśnierczyk P.: Single nucleotide polymorphisms of the *ERAP1* gene and risk of NSCLC: a comparison of genetically distant populations, Chinese and Caucasian. *Arch Immunol Ther Exp*, 2016,. 64, 117-122 suppl. 1 **IF - 2,464 (25 pkt. Min.)**

140. Ząbek A., Świerkot J., Malak A., Zawadzka I., Deja S., Bogunia-Kubik K., Młynarz P.: Application of the 1H NMR-based serum metabolomic studies for monitoring of patients with rheumatoid arthritis. *J Pharm Biomed Anal*, 2016; 117: 544-50. **IF – 3,169 (35 pkt. Min.)**

141. Zerka A, Rydzak J, Lass A, Szostakowska B, Nahorski W, Wroczyńska A, Myjak P, Krotkiewski H, Jaśkiewicz E.: Studies on immunogenicity and antigenicity of baculovirus-expressed binding region of *Plasmodium falciparum* EBA-140 merozoite ligand. *Arch Immunol Ther Exp*, 2016, 64 (2), 149-156 **IF - 2,464 (25 pkt. Min.)**

142. Zerka A., Olechwier A., Rydzak J., Kaczmarek R., Jaśkiewicz E.: Baculovirus-expressed *Plasmodium reichenowi* EBA-140 merozoite ligand is host specific. *Parasitol. Int*, 2016, 65, 708-714 **IF - 1,860 (25 pkt. Min.)**

143. Ziomkiewicz A, Sancilio A, Galbarczyk A, Klimek M, Jasienska G, Bribiescas RG.: Evidence for the Cost of Reproduction in Humans: High Lifetime Reproductive Effort Is Associated with Greater Oxidative Stress in Post-Menopausal Women. *PLoS One*. 2016 Jan 13;11(1):e0145753. doi: 10.1371/journal.pone.0145753. **IF – 3,057 (40 pkt. Min.)**

144. Ziemska J, Guśpiel A, Jarosz J, Nasulewicz-Goldeman A, Wietrzyk J, Kawęcki R, Pypowski K, Jarończyk M, Solecka J.:. Molecular docking studies, biological and toxicity evaluation of dihydroisoquinoline derivatives as potential anticancer agents. *Bioorgan Med Chem*. 2016, 24(21): 5302-5314. **IF – 2,923 (30 pkt. Min.)**

145. Zwolińska K, Błachowicz O, Tomczyk T, Knysz B, Gąsiorowski J, Zalewska M, Orzechowska B.U, Sochocka M, Piasecki E.: The effect of killer cell immunoglobulin-like receptor (KIR) genes on susceptibility to HIV-1 infection in the Polish population. Immunogenetics, 2016, 68: 327-337 **IF – 2,303 (20 pkt. Min.)**

146. Żaczek M,. Łusiak-Szelachowska M., Jończyk-Matysiak E., Weber-Dąbrowska B., Międzybrodzki R., Owczarek B., Kopciuch A., Fortuna W., Rogóż P., Górski A. Antibody production in response to staphylococcal MS-1 phage cocktail in patients undergoing phage therapy. *Frontiers in Microbiology*, 2016, vol 7, art. 1681 **IF – 4,165 (35 pkt. Min.)**

147. Żądzińska, E., Kozieł, S., Borowska-Strugińska, B., Rosset, I., Sitek, A., Lorkiewicz, W.: 2016. Parental smoking during pregnancy shortens offspring's legs. *HOMO – Journal of Comparative Human Biology,* DOI: 10.1016/j.jchb.2016.06.002 **IF – 0,579 (25 pkt. Min.)**

Publications from the journals not included in JCR database:

1. AbdelHamid H.F, Wietrzyk J, Pawlik A, Soliman AM.: Evaluation of In vitro Antiproliferative Potency and In vivo Induced Biochemical Parameters of Curcin from Jatropha curcas Seed in Comparison to Doxorubicin and Cisplatin. *Res. J. Pharm., Biol. Chem. Sci.* 2016, 7 (1): 719-738.
2. Bawiec A., Pawęska K, Jarząb A.: Changes in the microbial composition of municipal wastewater treated in biological processes. *Journal of Ecological Engineering*; 2016; 17(3),41–46 DOI: 10.12911/22998993/63316\ **(12 pkt. Min.)**
3. Borysławski, K., Kozieł, S.: Secular trend in body weight and length of children aged 0-2 years. Longitudinal study of five consecutive birth cohorts between 1964-2003 from Wrocław, Poland. *Anthropological Review,* 2016, 79(3): 301-310 **(15 pkt. Min.)**
4. Brodzik K, Augustynowicz E, Korzeniowska-Kowal A, Lutyńska A.: Zastosowanie metody MALDI-TOF do identyfikacji szczepów *Clostridium perfringens*. *Med. Dośw. Mikrobiol.,* 2016, 68: 13 – 21; **(7 pkt Min.)**
5. Ciołek L, Olszyna A, Zaczyńska E, Czarny A, Biernat M.: In vitro studies of antibacterial activity of sol-gel bioglasses containing Mg, Sr and Au. *Engineering of Biomaterials*. 2016; 134, 25-30. ISSN 1429-7248 Keywords: bioglass, sol-gel methods, cytotoxicity, antibacterial activity. **(7 pkt. Min.)**
6. Kozieł, S., Gomula, A., Nowak-Szczepanska, N. 2016.: The association between social factor and body length proportions in Poland schoolchildren from Lower Silesia. *Anthropological Review*, 79(4): 397-410 **(15 pkt. Min.)**
7. Njamkepo E, Fawal N, Tran-Dien A, Hawkey J, Strockbine N, Jenkins C, Talukder KA, Bercion R, Kuleshov K, Kolínská R, Russell JE, Kaftyreva L, Accou-Demartin M, Karas A, Vandenberg O, Mather AE, Mason CJ, Page AJ, Ramamurthy T, Bizet C, Gamian A, Carle I, Sow AG, Bouchier C, Wester AL, Lejay-Collin M, Fonkoua MC, Le Hello S, Blaser MJ, Jernberg C, Ruckly C, Mérens A, Page AL, Aslett M, Roggentin P, Fruth A, Denamur E, Venkatesan M, Bercovier H, Bodhidatta L, Chiou CS, Clermont D, Colonna B, Egorova S, Pazhani GP, Ezernitchi AV, Guigon G, Harris SR, Izumiya H, Korzeniowska-Kowal A, Lutyńska A, Gouali M, Grimont F, Langendorf C, Marejková M, Peterson LA, Perez-Perez G, Ngandjio A, Podkolzin A, Souche E, Makarova M, Shipulin GA, Ye C, Žemličková H, Herpay M, Grimont PA, Parkhill J, Sansonetti P, Holt KE, Brisse S, Thomson NR, Weill FX.: Erratum: Global phylogeography and evolutionary history of Shigella dysenteriae type 1. *Nat Microbiol*. 2016, Oct 3;1(11):16209.
8. Zwolińska K, Fleischer-Stępniewska K, Knysz B, Błachowicz O, Piasecki E.: Genetic diagnosis of seronegative (HIV-) partner of female patient with AIDS in the context of HIV transmission. *HIV AIDS Rev*., 2016; 15: 97-100 **(11 pkt. Min.)**

Books:

1. Kozieł S. (Redakcja naukowa): „*Wielowymiarowość badań biodemograficznych i związki z innymi naukami"*. Wydawnictwo Uniwersytetu Łódzkiego, Łódź. Organizatorzy: Komitet Biologii Organizmalnej PAN i Polskie Towarzystwo Antropologiczne, 2016, ISBN 978-83-8088-439-7, e-ISBN 978-83-8088-440-3

2. Kozieł S., Chakraborty R., Bose K.: Biological Implications of Human Mobility. NOVA Science Publishers Inc. New York, 2016, 1-166 str. ISBN: 978-1-63485-644-7

Chapters of books:

1. Bugdol, M. D., Bugdol, M. N., Lipowicz, A. M., Mitas, A. W., Bieńkowska, M. J., Wijata, A. M., Danel, D.: Longitudinal Voice Study (LoVoiS) Methodology and Preliminary Research Results. In: „ *Information Technologies in Medicin"*. 2016, 389–397. DOI: 10.1007/978-3-319-39796-2_32
2. Bełcik M, Piekarska K, Czarny A. Zaczyńska E.: Test kometowy jako metoda badań genotoksyczności pyłowych zanieczyszczeń powietrza atmosferycznego. W: *„Powietrze atmosferyczne: jakość - zagrożenia – ochrona".* Praca zbiorowa pod redakcją Kazimierza Gaja i Józefa Kuropki. Oficyna Wydawnicza Politechniki Wrocławskiej, ISBN 978-83-7493-942-3, Wrocław, 2016, s. 30-39
3. Chakraborty, R., Koziel, S., Bose, K.: Is the effect of age on diastolic blood pressure modified by the age at migration? A study among the adult male slum dwellers in Eastern India. W: *„Biological Implication of Human Mobility*". W: S. Koziel, R. Chakraborty, K. Bose (eds.). NOVA Science Publishing, New York, ISBN 978-1-63485-644-7, 2016, pp. 27-40.
4. Chełmońska-Soyta A, Kędzierska A.: Przyczyny wczesnej utraty ciąży u bydła. W: monografii *„Noworodek a środowisko",* Wrocław, 2016. str. 49-59 ISBN 978-83-927907-6-1
5. Frydecka I.: Czerwienica prawdziwa. W: „*Interna"* A. Szczeklika 2016, wydanie w oprawie miękkiej ISBN 978-83-7430-90-0; wydanie w oprawie twardej ISBN 978-83-7430-461-0 Medycyna Praktyczna, Kraków, 2016, str. 1739-1742
6. Frydecka I.: Nadpłytkowość samoistna. W: *„Interna"* A. Szczeklika 2016, wydanie w oprawie miękkiej ISBN 978-83-7430-489-4; wydanie w oprawie twardej ISBN 978-83-7430-461-0 Medycyna Praktyczna, Kraków 2016, str. 1742-1745
7. Frydecka I.: Pierwotne włóknienie szpiku. W: *„Interna"* A. Szczeklika 2016, wydanie w oprawie miękkiej ISBN 978-83-7430-490-0; wydanie w oprawie twardej ISBN 978-83-7430-461-0 Medycyna Praktyczna, Kraków 2016, str. 1745-1748
8. Frydecka I.: Czerwienica prawdziwa. W: *„Interna"* A. Szczeklika. mały podręcznik 2016/2017 wydanie VIII w oprawie miękkiej ISBN 978-83-7430-491-7; Medycyna Praktyczna, Kraków 2016, str. 933-936
9. Frydecka I.: Nadpłytkowość samoistna. W: *„Interna"* A. Szczeklika. mały podręcznik 2016/2017 wydanie VII w oprawie miękkiej ISBN 978-83-7430-461-7; Medycyna Praktyczna, Kraków 2016, str.933-936
10. Frydecka I.: Samoistne włóknienie szpiku. W: *„Interna"* A. Szczeklika. wydanie VIII w oprawie miękkiej ISBN 978-83-7430-490-0; Medycyna Praktyczna, Kraków 2016, str. 938-940
11. Koziel, S., Gomula, A.: Does social mobility of fathers affect age at menarche in daughters?. W: *"Biological Implication of Human Mobility*". W: S. Koziel, R. Chakraborty, K. Bose (eds.) NOVA Science Publishing, New York, ISBN 978-1-63485-644-7, pp. 2016, 121-136.
12. Lipowicz, A. M., Mitas, A. W., Bugdol, M. D., Bugdol, M. N., Bieńkowska, M. J., Wijata, A. M., Danel, D. 2016. Pre and Post Menarche—Girls' Bodies Parameters Comparison. In*:* „*Information Technologies in Medicine",* 2016, 399–408. DOI: 10.1007/978-3-319-39796-2_33
13. Łusiak-Szelachowska M., Żaczek M., Weber-Dąbrowska B., Kłak M., Międzybrodzki R., Fortuna W., Rogóż P., Szufnarowski K., Jończyk-Matysiak E., Górski A.: Antiphage activity of sera from patients receiving staphylococcal phage preparations. In: *Microbes in the spotlight: recent progress in the understanding of beneficial and harmful microorganisms*. Eds. A. Méndez-Vilas, BrownWalker Press, 2016, pp. 245-249. ISBN-10:1627346120, ISBN-13:9781627346122.

14. Pietkiewicz B, Boratyński J, Gamian A, Leszek J.: Stres oksydacyjny, końcowe produkty zaawansowanej glikacji i zaburzenia naczyniowe w chorobie Alzheimera. W: „Choroba Alzheimera - wybrane zagadnienia biologiczne i kliniczne", red. Jerzy Leszek, Wydawnictwo Continuo, 2016, 119-132 ISBN: 978-83-62182-57-2
15. Sochocka M: Stan zapalny w mózgu (Neuroimflammation). W: *„Choroba Alzheimera – wybrane zagadnienia biologiczne i kliniczne*", red.: Jerzy Leszek, Wydawnictwo Continuo, 2016, 133-147 ISBN 978-83-62182-57-2
16. Zabłocka A, Janusz M.: Zaburzenia szlaków sygnałowych w procesach neurodegeneracyjnych – rola kinaz białkowych aktywowanych mitogenami (MAP) *w* książce: *Choroba Alzheimera – wybrane zagadnienia biologiczne i kliniczne",* red. J. Leszek, 2016, str: 185-203, wyd. Continuo ISBN 978-83-62182-57-2

Popularized scientific article:

Myga-Nowak M, Godela A, Lewańska M, Głąb T, Boratyński J.: *The Academy of Young Inventor*, Akademia Młodych Wynalazców – inicjatywa poszerzająca horyzonty postrzegania świata. *Edukacja Biologiczna i Środowiskowa*", 2016/3, pp. 130-133