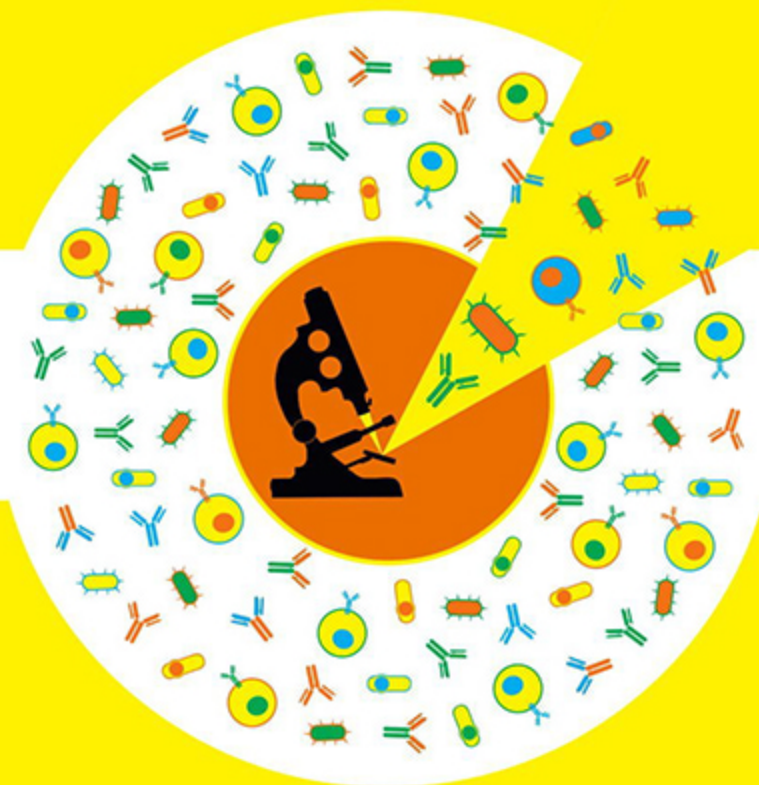


III international scientific conference

**MICROBIOLOGY AND IMMUNOLOGY -
THE DEVELOPMENT OUTLOOK
IN THE 21st CENTURY**



ABSTRACT BOOK

Kyiv
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The authors are responsible for the trustworthiness of scientific results and for the text of abstracts.

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PLENARY SESSION

Budzanivska I.G.

BACTERIAL VIRUSES IN THE HUMAN BODY

Taras Shevchenko National University of Kyiv, Kyiv, Ukraine

fitovirus@yandex.ru

Viruses are essential members of the human microbiome. Many of the viruses comprising the human virome have been identified as bacteriophage. This was only revealed in recent metagenomic studies. The influence of the presence of phages in humans has yet to be evaluated; but as in marine environments, a clear role in the regulation of bacterial populations could be envisaged, that might have an impact on human health. Moreover, phages are excellent vehicles of genetic transfer, and they contribute to the evolution of bacterial cells in the human body by spreading and acquiring DNA horizontally. The abundance of phages in the human body does not pass unnoticed and the immune system reacts to them, although it is not clear to what extent. The close association of phage with their cellular hosts suggests their communities may change in response to shifts in bacterial community membership.

Boyko N.V.

HUMAN MICROBIOME AS A KEY INSTRUMENT IN P4 MEDICINE AND IN PREVENTION OF INFECTIOUS AND NONCOMMUNICABLE DISEASES

R&D Centre of Molecular Microbiology and Mucosal Immunology, Uzhhorod National University, Ukraine

nadiya.boyko@gmail.com

Personalized nutrition, due to its direct effects on the modulation and maintenance of the human microbiome, is the key instrument for the early diagnostic and prevention of “food-relevant” diseases and their comorbidities. NCD prevention and treatment can be effectively managed when nutritional recommendations are applied on an age-group-specific and patient-centered basis. The new trend of “functional food” today can be defined as personalized, safe and tasty products, aimed at meeting individual requirements in nutrition while simultaneously maintaining microbial balance, variety and functions. Furthermore, pharmabiotics such as pre-, pro and synbiotics and their

metabolites should be also individually prescribed based on the different nosology as well as on the micro-immuno-biome characteristics of individuals.

The combination of (i) in vitro-based experiments; (ii) studies in germ-free mice and in transgenic mouse models of chronic and acute inflammation; (iii) clinical data of randomized trials and (iv) limited diet / nutrition studies have provided a number of very promising practical solutions for enabling the application of personalized nutrition. However, the practical use of bioinformatics to create algorithm(s) for the calculation of personalized nutrition is proving highly challenging due to the huge variety and diversity of existent data in this field. We are currently faced with the absence of unified approaches for the harmonization, evaluation, verification and proper exploitation of data with the aim of practical implementation in preventive, predictive and personalized medicine.

Our aim is exploitation of newly obtained molecular mechanisms and epigenetic tools in personalised medicine; we have been focused on providing evidence based data to modulate host immune response locally by reconstitution of composition of commensal microbes / individual microbiome and in such a way to develop and implement personalised diets and/or pharmabiotics for patient-centred prevention of the variety of non-communicable diseases with inflammation as a common trigger factor of initiation or infections caused by multi resistant strains of microbes.

Dons'koi B.V.

ACCENTUATED PHENOTYPE OF ENDOMETRIAL LYMPHOCYTES IN PATIENTS WITH REPEATED IMPLANTATION FAILURES ON PGD TESTED EMBRYOS

Institute of Pediatrics, Obstetrics and Gynecology, National Academy of Medical Sciences of Ukraine, Kyiv, Ukraine

boris_donskoy@ukr.net

Problems: Wide introduction of pre-implantation embryo diagnostics returns us back for studying significance of endometrial receptivity and local immune environment for successful implantation.

Method: We worked out the original methods for lymphocyte isolation from endometrium. We study phenotype of isolated endometrial (EL) and peripheral blood lymphocytes (BL) from healthy women (egg donors ED) on day of egg retrieval OV (n=24) and implantation window (IW) P6-P8 (n=33). IW samples were taken in stimulation (SC) (n=24) and post-stimulation cycle (PostS) (n=9). Similarly, we studied 13 samples on SC from patients with RIF (at least 1 failure

on PGD tested embryo). Lymphocyte phenotype was analyzed by flow cytometry (T and NK expression of CD4/CD8/CD16/CD158/HLADR/CD69/CD62L/CD335 were studied).

Results: ED groups formed conditionally “normal” EL subset levels for ovulation and IW. RIF endometrium consist same %NK but NK and T cell express more HLADR than ED. More than half of RIF have this levels out of “normal”. Significant part of RIF patients have “abnormal” expression levels of CD8, CD16, CD56 and CD56density on endometrial NK.

Decreased of CD56++ was the same as in ED in postS cycles but in contrast to ED where CD56dim were predominantly CD16neg, in RIF CD56dim were generally CD16+. Unlikely to blood NK these endometrial population (CD3-CD56dimCD16+) was extremely HLADR+.

Another atypical subset in RIF endometrium was CD3+CD16+ and CD3+CD335+ (both wasn't registered in ED endometrium but in part of RIF population these cells were present in generous frequency and inter-correlate. Abnormal EL phenotypes in RIF patients generally weren't reflected in PB. However, in PB from RIF patients significantly more often elevated levels of CD3CD158, CD3CD4HLADR, and NK lymphocytes were detected.

Conclusion: Endometrial lymphocyte phenotypes reflect receptivity as well as hormonal environment. Patient with RIF on PGD tested embryo typically have non-receptive endometrium that characterized by accentuated levels in lymphocyte subsets.

Garmasheva I.L.

COMPARATIVE CHARACTERISTIC OF LACTIC ACID BACTERIA PROPERTIES ISOLATED FROM TRADITIONAL FERMENTED FOODS

Zabolotny Institute of Microbiology and Virology National Academy of Sciences of Ukraine
Inna.garmasheva@gmail.com

Lactic acid bacteria (LAB) play an important role in the nature and have a great practical importance in human life. Traditional fermented foods, produced by spontaneous fermentation of milk and vegetables raw materials, are an inexhaustible source of biologically active strains with beneficial properties, that allow them to be used in the food industry and medicine. In the samples of fermented products investigated by us, the frequency of isolation of different LAB genera strains depended on the type of product, the source of milk, and the type of vegetable raw materials. Correlation of a number of physiological, biochemical and chemotaxonomic features of LAB strains with a source of isolation was

established, which significantly complicates phenotypic identification. Features of acid-producing and antagonistic activities of LAB strains depending on the taxonomic position and source of isolation was characterized. Among the samples of dairy products studied, the frequency isolation of enterococci was 60% of the total number of LAB and this causes some concern. An analysis of the antibiotic resistance of enterococci strains showed that resistance to benzylpenicillin, vancomycin, norfloxacin, rifampicin, doxycycline, and gatifloxacin correlates as with species and with source of isolation. Thus, artisanal fermented products are a source of biologically active LAB strains, but at the same time, dairy products can serve as a reservoir of virulent, enterococci strains with resistance to multiple antibiotics that are potentially dangerous to human health, which requires increased attention to this problem.

Havrylyuk A.M.¹, Chopyak V.V.¹, Kurpisz M.M.²

**IMMUNE REACTIVITY DISORDERS IN INFERTILE MEN WITH
REPRODUCTIVE SYSTEM ABNORMALITIES, SYSTEMIC AUTOIMMUNE,
CHRONIC INFLAMMATORY AND OTHERS DISEASES**

¹Medical University named Danylo Halytzhky, Lviv, Ukraine;

²Institute of Human Genetic of Polish Academy of Science, Poznan, Poland

ahavrylyuk@meta.ua

Introduction. Immune system plays an important role in male reproductive function, therefore her disorders are associated with formation of immune-dependent infertility.

Aim. The aim of this work was to determine local (in semen) and systemic (in peripheral blood) immunologic mechanisms in the formation of immune-dependent male infertility using clinical material.

Materials and methods. We were examined 71 men with with systemic inflammatory diseases of the connective tissue (SIDCT) – 18, somatic diseases on the background of chronic inflammation – 10, varicocele – 22, idiopathic infertility – 13, men, whose wives had early miscarriages – 8. For this patients was performed: spermiogram, antispermal antibodies (ASA), cytokines IFN- γ , TNF- α , IL-1 β , IL-6, IL-10, IL-18 and MDA; immunophenotyping of populations and subpopulations of lymphocytes; autoantibodies of the classes IgG and IgM to antiphospholipid and MPO, MDA. **Results.** In men with infertility and SIDCT the factors, affecting fertility, are: ASA of IgA class in seminal fluid (Tt); increased number of lymphocytes CD4+CD25+ and CD4+CD25+ T-cells; increased levels of TNF- α , IL-6, IL-18 in blood serum; decreased concentration of TGF- β 1 and IL-

1 β in seminal fluid; increased level of IL-18 in seminal fluid; increased level of antinuclear and antibodies of the classes IgG and IgM to phospholipids and β 2-GP-1, antibodies to MPO; increased level of MDA in blood and seminal fluid. In patients with varicocele we detected the changes of the following parameters: increased levels of IFN- γ and IL-18 in blood; TNF- α and TGFP- β 1 in seminal fluid; increased level of MDA in blood. **Conclusions.** We evaluated that changes in sexual system, causing sub- and infertility are initiated by different immune factors – triggers of infertility. Oxidation stress and apoptosis are the main mechanisms of infertility in groups with varicocele and somatic diseases on the background of chronic inflammation. The changes of lymphocyte and autoantibodies are the important mechanisms in SIDCT, idiopathic infertility. Generally cytokine disbalance, oxidative stress, apoptosis play a key role in formation of immune-dependent male infertility.

Hnatush S.O.

ABILITY TO EXOELECTROGENESIS OF BACTERIA, WHICH ARE ISOLATED FROM TECHNOGENICALLY TRANSFORMED ENVIRONMENTS

Ivan Franko National University of Lviv, Lviv, Ukraine

gnatuk88@ukr.net

Metabolic activity of exoelectrogenic microorganisms in bioelectrochemical systems, in particular, in microbial fuel cells (MFC), provides electric current generation, which is combined with anaerobic organic matter oxidation. In MFC oxidation of organic compounds occurs with participation of microorganisms in anode chamber under anaerobic conditions where anode is used as a sole electron acceptor. Anode and cathode chambers are separated by proton selective membrane, which causes the transfer of protons and prevents oxygen access. Cell exoelectrogenesis in anode chamber causes electric current generation. MFC are perspective for waste water treatment from organic contamination. For bioremediation of contaminated territories it is importantly to use strains of bacteria, which are isolated from technogenically transformed environments and therefore are resistant to high concentrations of hydrogen sulfide, sulfates, heavy metal salts etc. Strains-exoelectrogens were isolated from Yavoriv Lake, which was created on the territory of the sulfur career, and infiltrates of Lviv solid waste landfill (Lviv region, Ukraine).

Electron donors have been selected for electric current generation by *Desulfuromonas acetoxidans* IMV B-7384 in a MFC. It was established, that power density of MFC under the usage of electron donors, which metabolism is

accompanied by the release of reduced equivalents, were higher, in comparison with electron donors, conversion of which is unrelated with formation of reduced equivalents. Power density of MFC with application of *Chlorobium limicola* IMV K-8 as anolyte during seven days of cultivation were 1–1.2 W/m². Purple photosynthetic bacteria *Rhodospseudomonas yavorovii* IMV B-7620 were also able to eclectic current generation.

Isolated strains formed electric current in MFC, using the waste water of yeast factory and infiltrates of the Lviv solid waste landfill as a substrate. Power density of MFC with *R. yavorovii* IMV B-7620 and waste water of yeast factory during seven days of cultivation were 0.8–2.1 W/m². Under the usage of infiltrates of Lviv solid waste landfill by *D. acetoxidans* IMV B-7384 bacteria power density of MFC were 0.7–2.1 W/m².

Thus, bacteria, which were isolated from technogenically transformed territories, are perspective for MFC construction that opens wide aspects for development new alternative energy sources.

Khranovska N.M., Skachkova O.V., Gorbach O.I., Inomistova M.V.

**IMMUNOLOGY OF MALIGNANT NEOPLASMS: FROM CONCEPTUAL
ASPECTS TO THE IMMUNOTHERAPY PRACTICE**

National Cancer Institute, Kyiv, Ukraine

nkhranovska@ukr.net

Studies in the field of molecular tumor immunology and the possibilities of the immunological response modulation have made significant progress. Although, the place of immunotherapy in the treatment of malignant neoplasms is not defined definitively. One of the hypotheses that explain this fact is the notion of the significant role of the immune system in the course of malignant diseases and its possible insolvency in certain stages of the tumor progression. The breakthrough in the cancer immunotherapy is associated with an understanding of the mechanisms of interaction between the tumor and the immune system, features of T-cell regulation. Currently, such approaches to tumor immunotherapy are being used or under development: antitumor vaccines, including based on dendritic cells, monoclonal antibody, cytokines, checkpoint inhibitors, activated lymphocytes, genetically modified T-lymphocytes (CAR- T).

National Cancer Institute of Ukraine has elaborated novel DCs technology which is characterized by two cycles of innovation: 1. DCs activation and maturation with IFN- α plus toll-like receptor agonist and 2. DCs loading with mechanically modified microparticles of tumor cells. High therapeutic

effectiveness of this technology has been demonstrated in pre-clinical and clinical settings. In experimental studies found that DCs based vaccine therapy is an effective method for metastasis burden decreasing and primary tumor growth inhibition: index of metastasis inhibition was 66-95% in mice with Lewis lung carcinoma after removal of the primary tumor and 99% in mice with B16 melanoma. DC-vaccine therapy contributes significantly to improving the results of non small cell lung cancer treatment. Thus, during the 5-year follow-up period DC based vaccine increased the overall survival of patients by 25% (HR 0.47; 95: CI = 0.9-0.74), disease-free period – by 26% (HR 0.38; 95: CI = 0.24-0.61).

Thus, cancer immunotherapy — treatments that harness and enhance the powers of the immune system to fight cancer, represents the most promising new cancer treatment approach.

Kondratiuk T.O.

MICROORGANISMS DESTRUCTORS OF WALL PAINTING IN THE UNESCO WORLD HERITAGE SITE - ST. SOPHIA CATHEDRAL MUSEUMS, XI CENTURY, KYIV

Taras Shevchenko National University of Kyiv, Educational and Scientific Center "Institute of Biology and Medicine"

takbiofak@ukr.net

Microorganisms accepted to be among the major destructors of products and materials. Microscopic fungi destructors can pose a threat to human health (mycoallergy, mycosis, syndrome of ill rooms, emergence of hazardous emergencies due to deterioration of quality and violation of operational characteristics of equipment etc). The negative consequences of biodestruction are significant economic losses and the social aspect of harm - the loss of historical and cultural monuments. Preservation of the monument of the UNESCO World Heritage St. Sophia Cathedral Museum, XI century. "Sofia Kievskaya" National reserve is an actual task today. To ascertain the role of microorganisms as a component in damaging the unique wall painting in St. Sophia Cathedral Museum, identification of the main causes that contribute to the occurrence and development of these lesions; characteristic of biocidal effects on microorganisms test cultures was the purpose of this work. Samples of damaged wall painting, air samples, dust and samples of damaged walls in the basement of the cathedral were materials of research. It was established that damage to wall paintings in the St. Sophia Cathedral is due to the complex of microorganisms, the main component of which are xerophilic microscopic fungi of the genera *Emericella*,

Eurotium, and *Aspergillus*. Microscopic fungi of the genera *Penicillium*, *Cladosporium*, *Rhodotorula*, actinomycetes, and spore-forming bacteria of the genus *Bacillus* were also isolated. The periodic condensation moistening of the walls of the cathedral (due to disturbances of the temperature-humidity regime inside and the waterproofing of the walls of the building from the outside); presence of sources of airborne contamination of microorganisms (destroyed walls of basements, significant dust contamination in ventilation ducts) found to be the main causes of the occurrence and development of wall painting in St. Sophia Cathedral. The species composition of microscopic fungi and their quantitative indices in air and dust samples poses a threat to unique wall painting and human health. The low stability of test cultures of fungi to the influence of investigated biocides in various concentrations was established. An enhanced synthesis of pigments by fungi in response to the influence of biocides has been observed. The recommendations for improvement of situation were given based on results of work obtained.

Korotetskiy I.S.¹, Shilov S.V.¹, Shvidko S.V.¹, Jumagaziyeva A.B.¹, Suldina N.A.¹, Korotetskaya N.V.¹, Sabina K.T.¹, Ilin A.I.¹, Reva O.N.²

**DRUG RESISTANCE REVERSION: GETTING INSIGHT INTO MECHANISMS
AND PROSPECTS OF CLINICAL APPLICATION**

¹JSC Scientific Center for Anti-infectious Drugs, 75V al-Farabi Ave., Almaty, 050060, Kazakhstan;

²Department of Biochemistry, Centre for Bioinformatics and Computational Biology, University of Pretoria, Pretoria, South Africa

laeda81@gmail.com

Abstract: The emergence and wide distribution of antibiotic resistant pathogens is a serious threat to the public health. The development of new antibiotics experiences a deep crisis due to increased cost of production and fast emerging of resistant pathogens. Drug induced reversion of antibiotic resistance is a promising way to combat multidrug resistant infections. However, lack of knowledge of mechanisms of this phenomenon impedes employing this approach in medicinal therapies. FS-1 is a nanomolecular iodine containing complex, which induces a reversion of drug resistant bacteria to drug sensitive phenotypes. The effect has been studied on multidrug resistant strains of *Mycobacterium tuberculosis* [1], *Staphylococcus aureus* [2] and *Escherichia coli* by using laboratory models and modern techniques of DNA and RNA sequencing. It was found that FS-1 causes an oxidative stress on bacteria that increases the fitness

cost of the drug resistance. Moreover, the cultivation of bacteria with sub-lethal concentration of FS-1 leads to a significant long-lasting disruption of gene regulation that induces the drug resistance reversion even after removal of FS-1 from the medium. Variant calling in genome sequences of pathogens revealed no mutations which could explain the aberrant gene regulation. That raised a hypothesis of epigenetic mechanisms to be involved. Indeed, PacBio sequencing revealed patches of significant DNA modification resulted from methylation and possibly from halogenation of nucleotides in bacterial chromosomes. This work demonstrated the epigenetic nature of the long lasting drug resistance reversion induced by FS-1. Molecular mechanisms of the resistance reversion were studied by comparison of gene expression patterns and identification of key transcriptional factors involved in gene regulation under the influence of FS-1, which eventually caused aberrant epigenetic modifications of bacterial chromosomes and decreased the viability of microorganisms. This work will aid in improving of regimens of the clinical application of FS-1 and guide the design of new drugs.

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Lisyaniy N.I.

IMMUNE CONVERSATION OF BRAIN CELLS

The State Institution Romodanov Neurosurgery Institute, National Academy of Medical Sciences of Ukraine, Kyiv, Ukraine

nimun.neuro@gmail.com

Today, the classical theory of immunological isolation of the brain has been replaced by the idea of the presence of an immune system in the brain that is able to function autonomously or together with the reactions of systemic immunity. The autonomous immune system of the brain performs at least 3 different functions: anti-infection protection and support of antigenic brain

homeostasis, neurodegeneration and demyelination, neuroregeneration and repair of damaged cells and brain tissues. These functions are cooperatively performed by microglial cells, neurons and astrocytes, which express TLR receptors, C-200 molecules, Fas-ligands, adenosine, chemokines and cytokines. In physiological and pathological conditions, there is a kind of immunological conversation between these cells using immunoregulatory factors. A deep knowledge of the mechanisms of such a conversation gives hope for a possible correction of these reactions in future.

Tkachenko Y.V.¹, Vorobyova G.M.¹, Yakovenko L.F.², Yemets I.M.¹

**CARDIAC REPAIR USES AUTOLOGOUS UMBILICAL CORD BLOOD IN
NEWBORNS WITH CONGENITAL HEART DISEASE AS A NEW
BIOTECHNOLOGICAL APPROACH**

¹Department of bio tissue and reconstructive surgery, Ukrainian Children's Cardiac Center, Kyiv, Ukraine;

²Laboratory of molecular mechanisms of autoimmune processes, Institute of molecular biology and genetics, Kyiv, Ukraine

yani_t2008@ukr.net

About 5 thousands of children with critical congenital heart disease (CHD) born in Ukraine annually. In Ukraine firstly in the world suggested to use autologous umbilical cord blood (AUCB) in cardiac surgical CHD as a new biotechnological method. The present work addresses major issues regarding search markers of safety and effectiveness of AUCB in neonatal. We analyzed molecular interactions of the immune system's proteins mediated Toll-like receptor and activate immune cell responses newborns with CHD after transfusion AUCB. New properties cord blood were revealed. We studied of the role of anti-Hsp60 antibodies, Sgt1, cytokine profile, glial protein - S100 β - as a marker of cerebral ischemia, HIF-1 β within development of postoperative complications in neonates with CHD after AUCB or donor blood transfusion during cardiac surgery. We were the first who studied the level of anti-Hsp60 antibodies and anti-Sgt1 antibodies in pregnant women with prenatally diagnosed CHD of fetus and clinically healthy donors whose blood components used for transfusion to neonates with CHD. It was established statistically significant difference between the level of anti-Sgt1 antibodies and anti-Hsp60 antibodies in the serum of pregnant women and their newborns ($p=0.000001$). For understanding the possible immunoregulatory role of these antibodies we need to

investigate their biological activity. We found that anti-Hsp60 antibodies in circulating newborns after donor blood transfused recognize the protein Hsp60 on the surface of cardiomyocytes by the immunoblotting method. This study demonstrates the potential therapeutic effects of AUCB compared donor blood donated.

**Tolstanova G.M.¹, Holota Y.O.¹, Dovbynchuk T.V.¹, Chervinska T.M.¹,
Sergeychuk T.M.¹, Zakordonetz L.V.², Kramarev S.O.²**

**DO ANTIBIOTICS INCREASE THE ACCIDENTS OF INFLAMMATORY BOWEL
DISEASES? EXPERIMENTAL DATA AND CLINICAL OBSERVATIONS**

¹Taras Shevchenko National University of Kyiv, Kyiv, Ukraine;

²Bogomoletz National Medical University, Kyiv, Ukraine

gannatolstanova@knu.ua

The disturbance of complex interaction between symbiotic aerobic and anaerobic intestinal bacteria is one of the critical mechanisms of inflammatory bowel disease (IBD) pathogenesis. Accordingly recent epidemiological studies, prolonged antibiotic therapy significantly increases the risk of IBD development.

We found that administration of the cephalosporin antibiotic ceftriaxone (300 mg/kg, 14 days) induced the stable downregulation of SCFAs level. These changes were associated with decrease immunoreactivity of the FFA2 and FFA3 receptors in rats' colon mucosa. Ceftriaxone administration decreased the immunoreactivity for SMCT1 transporters of SCFA and increased the for MCT1 & MCT4 on enterocytes. These changes evoked a significant shift in colonic mucosal homeostasis; the disturbance of oxidant-antioxidant balance that induced reduction of protein SH-groups and activation of redox-sensitive transcription factors Egr-1, Sp-1, HIF1 α and ERK1/2 MAP kinase. Consequently, these changes provoked colonic mucus barrier dysfunction long-term after the antibiotic withdrawal; decrease the total concentration of mucus glycoproteins; changes in the mucins glycosylation. It was accompanied by increased activity of pro-inflammatory matrix metalloproteinase MMP-9 and decreased anti-inflammatory MMP-2 in colon mucosa. Mucus barrier defects were associated with increased permeability of colonic epithelium, increased bacterial translocation to the blood and associated with enhanced sensitivity to the experimental colitis. Thus, the disruption of colon barrier function is a significant long-term side-effect of antibiotic therapy, which increases susceptibility to the development of intestinal inflammation.

Experimental and clinical observations confirmed that the most effective probiotics treatment protocols to prevent long-term side-effect of antibiotic therapy on colonic function are: 1 - the application of multiprobiotic together with an antibiotic and for 10-14 days after antibiotic withdrawal; 2- the administration of multiprobiotic right after antibiotic withdrawal for 10-14 days.

Zubov D.A.

TISSUE REPAIR AS A GENUINE CROSS-TALK BETWEEN MULTIPOTENT MESENCHYMAL STROMAL CELLS (MSCs) AND INNATE IMMUNITY

State Institute of Genetic and Regenerative Medicine NAMSU, Kiev, Ukraine;

ILAYA® Medical Company, Kiev, Ukraine

zoubov77@yahoo.com

In our studies (2006-2010), it was shown that transplantation of cultured bone marrow-derived multipotent mesenchymal stromal cells (MSCs) for healing of long term fracture non-union and avascular osteonecrosis of the femoral head promotes the reparative osteogenesis processes in situ. Moreover, bone marrow-derived MSCs committed into the osteogenic direction are secreted in culture medium the multiple cytokines such as IL-1 β , IL-2, IL-4, IL-6, IL-8 and TNF α , with significantly increased production of osteoactive IL-6 (up to 276.5 pg/ml) and IL-8 (up to 1.06 ng/ml).

At present, the immunoregulatory properties of MSCs have been reflected in a mass of experimental and clinical studies dedicated to the functional characteristics of cultured MSCs ex vivo and to the fate of transplanted cells within the organism. Therefore, R. Waterman research team proposed in 2010 a new MSCs paradigm, consisting in the ability of these cells, depending on the received microenvironment signals, to polarize either to the pro-inflammatory (MSC1) or to the anti-inflammatory, or immunosuppressive (MSC2) phenotypes (like M1 and M2 macrophages paradigm).

Thus, it is known that cultured MSCs interact directly with the immune system of the recipient organism and are an important component of the reactions of the innate immune response. The innate immunity acts as a key partner in interaction with MSCs in the aspect of tissue repair and it is a mediator of some MSCs trophic functions. Also, MSCs are able to directly recognize the pathogen in the microenvironment and generate the antimicrobial response at the local and systemic levels, which leads to a significant reduction in bacterial load and subsequent animal survival in experimental sepsis models. MSCs promote wound healing through the recruitment of monocytes and macrophages with their further induced polarization from M1 to M2 macrophages. MSCs, monocytes and macrophages form together effective feedback loop mechanisms to prevent an

excessive inflammatory response in the wound and its resolution towards the realization of remodeling and reparative regeneration processes within affected tissues.

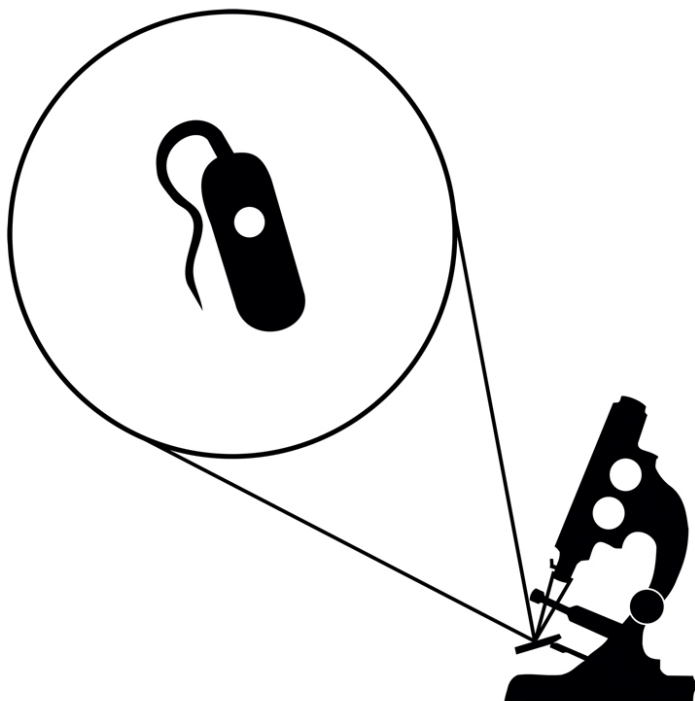
Umansky V.

TUMOR-INDUCED IMMUNOSUPPRESSION AND ITS OVERCOMING: NEW APPROACHES

German Cancer Research Center (DKFZ), Heidelberg, Germany
v.umansky@dkfz.de

Malignant tumors are characterized by fast progression and poor response to conventional therapies (such as chemo- and radiotherapy). Therefore, immunotherapeutic strategies could be helpful. However, their efficiency could be significantly reduced by the chronic inflammation represented by infiltrating leukocytes and soluble mediators that result in the immunosuppression in tumor microenvironment. Using ret transgenic mouse melanoma model, which mimics clinical situation in human melanoma, we demonstrated increased levels of chronic inflammatory factors and immunosuppressive cells (like myeloid-derived suppressor cells, MDSC) in melanoma lesions that correlated with reduced anti-tumor T cell reactivity and accelerated tumor progression. Inhibitors of MDSC activities were shown to mediate strong anti-tumor effects. In cancer patients, levels of serum inflammatory factors and frequencies of circulating immunosuppressive cells were increased that significantly correlated with a decreased progression free survival. Moreover, decreased MDSC frequency and immunosuppressive pattern correlated with the patients' response to immunotherapy. We suggest that inhibitors of immunosuppressive tumor microenvironment should be included in combined tumor immunotherapy to increase it efficiency.

SECTION



MICROBIOLOGY

Abdulina D.R.¹, Chuenko A.I.¹, Bondarenko A.I.², Dolyuk O.V.²

ENZYMATIC ACTIVITY OF *DESULFOVIBRIO DESULFURICANS* UCM B-11501, *DESULFOVIBRIO SP.* UCM B-11503 STRAINS IN THE PRESENCE OF RUBBER AND POLYMERIC MATERIALS

¹D.K. Zabolotny Institute of Microbiology and Virology, NAS of Ukraine;

²National University of Food Technologies

adara@ukr.net

The explosion of various polymeric compounds production leads to the introduction of materials into all areas of society. But one of the disadvantages of polymers is that they have long-term degradation period and as a waste is causing harm to environment. Aim of the study was the estimating of the sulfate-reducing bacteria enzymatic activity as a one of the criteria for utilization ability of the rubber and polymeric materials. Objects of the study were strains of sulfate-reducing bacteria (SRB) *Desulfovibrio desulfuricans* UCM B-11501 (DSM642), *Desulfovibrio sp.* UCM B-11503 (10). Bacteria cultivation was performed in the Posgate "B" liquid media at 28 C, 14 days. According to the variant of experiment into the media were dipped previously weighted and sterilized samples of rubber, foamed polyethylene (FPE) and ethylene vinyl acetate (EVA). Activity of the catalase measured by the reaction with the molybdate salts and hydrogen peroxide, activity of the lipase - in the reaction with the p-nitrophenylpalmitate, amount of the protein in the cultural liquid - by the Lowry's method. Enzymatic activity of *Desulfovibrio sp.* 10 and *D. desulfuricans* DSM642 were highest in the control variant (without any materials), specific catalase activity were 0.66 - 1,40 u/mg of protein and lipase activity 39.2 – 25.6 u/mg of protein, respectively. *Desulfovibrio sp.* 10 in variants with rubber and FPE catalase activity were decreased in 2.8 times (0.24 – 0.23 u/protein mg, respectively) and not changed with the EVA. *D. desulfuricans* DSM642 strain catalase activity had decreased in all variants – with rubber, PPE and EVA in 21.4%, 12.8% and 37.0%, respectively. Lipase activity of *Desulfovibrio sp.* 10 were decreased in 1.9 times in variant with rubber, with FPE were similar to control and increased in 34.5 u. with the EVA. Lipase activity of *D. desulfuricans* DSM642 with the rubber and FPE were increased in 16,07 and 21,9 u. and with EVA decreased in 8,55 u. Rubber and FPE were influenced by *D. desulfuricans* DSM642 and EVA – by *Desulfovibrio sp.* 10. Thus, we could conclude that lipase activity is representative for estimating of polymeric materials biodegradation by SRB.

Abdurashytov S., Egovtseva A.**DETECTION OF NONRIBOSOMAL PEPTIDE SYNTHETASE GENES IN THE STRAINS OF PHYTOPATHOGEN-ANTHAGONISTS AND THEIR EXPRESSION**

FSBSI Research Institute of Agriculture of Crimea, Simferopol, Crimea

asuleyman83@rambler.ru

Benefit microorganisms can serve as biocontrol agents of mass bacteriosis expansion. Currently, an active search and study of antagonists for various pathogens is conducted. One of the mechanisms of their antimicrobial action can be nonribosomal peptides (NRP) production (bacteriocins, antimicrobial peptides). The aim of the investigation was establish an antagonistic action of *Bacillus amyloliquefaciens* strains and analyze their NPR synthetase gene expression in interaction with *Clavibacter michiganensis*.

B. amyloliquefaciens 01-1 and *B. amyloliquefaciens* 01-2 is the strains of phytopathogens antagonists from the Crimean collection of beneficial microorganisms. *C. michiganensis* subsp. *michiganensis* 10₂ (provided by IMV NAS of Ukraine) – causative agent of tomato bacterial canker. The method of perpendicular strokes on fish-peptone agar (FPA) was used to determine the influence of the antagonists against the pathogen. The mechanism of antagonistic action was revealed by the detection of NPR synthetase genes in antagonists and their expression by PCR with specific primers to surfactin, bacilysin, macrolactin, bacillaene. The gene expression evaluation was quantified the PCR products, comparing the luminescence intensity of the DNA fragments separated in an agarose gel with the intensity of molecular-weight-standard via the image analysis program Total lab 2.1.

According to the data, *B. amyloliquefaciens* 01-2 formed 3.5 ± 0.1 mm zone of growth suppression of *C. michiganensis* 10₂ in their interaction on the FPA medium that shows its efficiency via the causative agent of bacteriosis. *B. amyloliquefaciens* 01-1 only stopped the growth of the test-microorganism. The availability of surfactin, bacilysin, bacillaene, macrolactin synthetase genes sites in *B. amyloliquefaciens* 01-1 and 01-2 was obtained. The antagonistic activity of *B. amyloliquefaciens* 01-2 on contact with phytopathogen can be achieved through the secretion of surfactin and bacilysin. The genes expression of this NPR increased at 21.0-22.7 times in comparison with the normal growth conditions of the microorganism.

Adejuwon A.O., Odeleye O.D., Olaiwole O.

**ANTIBIOTIC SUSCEPTIBILITY OF COLIFORM IDENTIFIED IN COW DUNG
FROM A MAJOR ABATTOIR IN IBADAN, NIGERIA**

Lead City University, Ibadan, Nigeria

ao_adejuwon@yahoo.ca

The present study aimed to estimate the extent of microbial contamination and antibiotic sensitivity of coliforms in cow dung samples from an abattoir in Ibadan, Nigeria. Samples of cow dung were collected from the different cows in an abattoir at Bodija, Ibadan, Nigeria. The microbial counts in the samples were enumerated using Nutrients Agar (NA), Eosine Methylene Blue agar (EMB) and Potato Dextrose Agar (PDA) for fungi. Eighteen different microorganisms were isolated from the cow dung samples. Of these, ten strains of bacteria were identified. These were *Micrococcus luteus*, *Staphylococcus aureus*, *Salmonella typhi*, *Staphylococcus epidermidis*, *Pseudomonas aureginosa*, *Arthrobacter sp*, *Bacillus subtilis*, *Cytophyaga sp*, *Streptomucetes sp*, *Citrobacter sp*. Six strains of moulds were also identified and were *Aspergillus fumigatus*, *Aspergillus nidulans*, *Botrytis cinerera*, *Fusarium sp*, *Trichoderma viriide* and *Aspergillus niger*. Two strains of yeast, *Trichospora sp* and *Endomyces sp*. were identified. These isolated organisms were characterized with their percentage (%) of occurrence as follows: Bacterial had the highest percentage (%) of occurrence (52%) followed by Fungi (44%) then Yeast (4%). The bacteria were sensitive to Nalixidic acid (NAL), Nitrofurantoin (NIT) Cotrimazole (COT), Tetracycline(TET), Gentamycin(GEN) and Amoxicillin(AML), Erythromycin (ERY), Chloxacillin (CXC), Streptomycin (STR). There was high resistance of the bacteria to Augmentin (AUG) and Ofloxacin. It was concluded in the study that different types of bacteria isolated from cow dung may have serious implications on human health. However, cow dung can be very useful in compost manure since it contains some important nutrients relevant in agriculture. The overuse of antibiotics (higher than the WHO limit) in treating cows may lead to antibiotic resistance in such cows. This is of global health concern. Therefore, intensive efforts must be initiated to identify and preserve all the indigenous breeds of cows for comparative chemical and microbiological analysis of dung with special reference to their agricultural, medicinal and nutritional significance. Adequate farm practice is also recommended.

Aghayeva E.M, Alasgarov Z.A, Gambarli I.D

ETIOLOGICAL STRUCTURE AND ANTIBIOTIC RESISTANCE OF
SALMONELLA - ACUTE INTESTINAL INFECTIOUS AGENTS IN AZERBAIJAN

Azerbaijan State University, Azerbaijan, Ganja

zahir.alaskarov@mail.ru

Objective. To study the etiological structure of acute intestinal infections (All), to identify the multi-drug resistance of *Salmonella* strains – All agents in Baku.

The multi-drug resistance of *Salmonella* strains is determined by R-plasmids capable of conjugation between bacteria in a population of the same species and bacteria belonging to the different species, genera and even families that provides selective advantages to the host cell.

So, multi-drug resistance of salmonella to laevomycetin, kanamycin, and ampicillin has been established. In recent years, there has been a spread among *Salmonella* strains resistant to cephalosporins of the third generation that is occurred to plasmid β -lactamases of extended spectrum.

Materials and methods. Identification of microorganisms isolated in All was carried out by the methods common in microbiology, as well as by Mari POC, based on the identification of multiplex PCR using a diarrheal panel, which includes the identification of 10 bacteria and 4 viruses. The sensitivity of the isolated salmonella strains to antibiotics was determined by the E test (Sweden) and Vitek-2 system.

Conclusions. It was isolated 140 strains – All agents, including 72 *Salmonella* strains, 48 diarrhea *E. coli* strains and 20 strains of other enterobacteria. Diarrheal strains are represented by enterotoxigenic (32) and enteroinvasive (6) *E. coli* and 10 enteropathogenic strains. The *Salmonella* strains belonged to the serovars of *S. enteritidis* – 51 strains and *S. typhimurium* – 18 strains, the rest are represented by single strains and belonging to other serovars. We have determined the resistance of salmonella to ampicillin (12%), chloramphenicol (8%), cotrimoxazole (4%), gentamicin (2%), cephalosporins (6%).

Thus, it has been determined the resistance of salmonella to clinically relevant antibiotics – beta-lactams and fluoroquinolones.

Agayeva E.M., Narimanov V.A., Seyidova G.M., Mammedova R.E., Huseynov R.M.

**METICILLIN RESISTANT *STAPHYLOCOCCUS AUREUS* AND COAGULASE
NEGATIVE STAPHYLOCOCCI AS PATHOGENS CAUSING INFLAMMATORY
SKIN DISEASES**

Azerbaijan Medical University, Baku, Azerbaijan

vnarimanov@gmail.com

Introduction. As a consequence of selective pressing of antibiotics applied in medicine, spread of antimicrobial resistance has become a global problem. Due to meticillin-resistant strains (MRSA) treatment of staphylococcal infections is an important problem. Staphylococci are among the leading causative agents of inflammatory skin diseases (ISD) in ambulatory and non-ambulatory patients. Thus, solution of this problem is of great importance.

Materials and methods. 78 *Staphylococcus* strains were detected after microbiological examination of patients with ISD. Identification of strains was performed using conventional procedures. Vitek 2 automated system and E-test (AB. Biodisc, Sweden) were used for antibiotic susceptibility testing.

Results. 78 *Staphylococcus spp.* strains were isolated from 125 patients with ISD. Identification on species level based on morphological and biological features of bacteria. Coagulase-negative staphylococci (CoNS) prevailed among isolated strains: *S.saprophyticus* – 18%; *S. hominis* – 4%; *S.warneri* – 2%; *S.epidermidis* – 16%; *S.lugdunensis* – 11%; *S.aureus* – 52% of isolated *Staphylococcus spp.* strains. Out of 65 *S.aureus* strains, 42 were meticillin-resistant, 23-meticillin susceptible. The majority of MRSA was obtained from ambulatory patients.

All 42 MRSA were susceptible to vancomycin, 82 %-to rifampin. Among coagulase-negative staphylococci 45% had resistance to oxacillin.

Conclusion. The obtained data proved that the leading causative agents in ISD were staphylococci. *S.aureus* was the main pathogen among *Staphylococcus spp.* Application of beta-lactame antibiotics can induce spread of MRSA strains and change epidemiological situation relating to MRSA strains. Continuous monitoring of etiological structure of ISD and detection of antibiotic resistant strains should be performed in order to develop effective and rational treatment schemes.

Akulenko I.¹, Korbush M.¹, Kornienko V.¹, Stepanova N.², Serhiychuk T.¹, Tolstanova G.¹

DYNAMIC CHANGES OF OXALATE-DEGRADING BACTERIA IN RATS COLON DURING ANTIBIOTICS-INDUCED DYSBIOSIS

¹Taras Shevchenko National University of Kyiv, Ukraine;

²State Institution «Institute of Nephrology of the National Academy of Medical Sciences», Kyiv, Ukraine

estee23@gmail.com

Oxalate-degrading bacteria (ODB), e.g. *Oxalobacter formigenes*, play a key role in the metabolism of oxalates. Individuals without ODB have increased risk of urinary stone diseases, e.g. hyperoxaluria. The **AIM** of the present study was to investigate the effect of combined administration of ampicillin (AMP) and metronidazole (MNZ) on number of ODB in fecal and mucosal biopsy of rat colon. **Methods.** The object of the study was fecal and mucosal microbiota of the colon of male Wistar rats (170-200g, n=22). To induce experimental dysbiosis rats were gavaged with AMP (75mg/kg, PJSC «Kievmedpreparat») and MNZ (50mg/kg, LTD Pharmaceutical company «Zdorovye») once a day for 3 days. Fecal microbiota was determined bacteriologically next day after antibiotics withdrawal, and in 18, 29 and 59 days. Colonic mucosal microbiota was determined in 59 days. ODB were identified by bacteriological culture method in the sodium oxalate reach medium, cultivated anaerobically at 37°C, 48 hr. **Results.** Combined administration of AMP/MNZ contributed to the development of dysbiosis 1-2 stage which was normalized from 18 to 59 days after AMP/MNZ withdrawal. Another picture was observed for ODB. Next day after cessation of antibiotics and in 18 days quantity of ODB in the feces remained at the level of control values (lg 6,75±0,48 CFU/g). At the 29 day of experiment the number of ODB was decreased on 2 orders (lg 4,37±0,80 CFU/g). The normalization of number of ODB was observed in feces in 59 days. But in the mucosal biopsy quantity of ODB in 59 days was on 2 order lower vs. control group of intact animals. Evaluating species diversity of ODB we found 8 morphotypes, among which the vast majority was Gram-positive bacilli and cocci, incl. spore-forming. Only one type of colonies was identified by microscopy like Gram-negative bacilli, which can be attributed to *O. formigenes*. In fecal biopsy the number of morphotypes didn't significantly change, in mucosal biopsy reduction in the variety of morphotypes and disappearance of Gram-negative non-spore-forming bacilli were noted. **Conclusion.** Antibiotic therapy may have a negative impact on the quantity of ODB even long-term after cessation.

Bakshaliyeva K.F., Muradov P.Z., Safarova A.Sh.

**THE ECOLOGICAL-BIOLOGICAL FEATURES OF TOXIGENIC FUNGI WHICH
SPREADS IN AZERBAIJAN**

Institute of Microbiology of Azerbaijan National Academy of Sciences, Baku, Azerbaijan
mpanah@mail.ru

As the result of the research, has been comprehensively investigated the soils in the ecologically different areas of Azerbaijan Republic, plant and aquatic ecosystems, as well as the mycobiota of materials used for variety of purposes (food, feed and medical), the number and species of composition of toxigenic in its formation, frequency of the prevalence, spreading laws, the ecobiology, also influence of anthropogenic factors to the spread of toxigenic fungi and to find the drugs for neutralization of harmful-activities of toxigenic fungi.

Referring to the analysis of samples taken during 2012-2017, 137 type of fungi have been isolated to pure culture and identified that. It was clear, 80 species from 130 belongs to toxigenic fungi, and in the result of phytotoxic activity relative to the plant determined that 19(50% higher) fungi have shown strong, 32(between 10-50%) medium and 25(10% less) weak phytotoxic activity.

It became clear that fungi contained in the formation of mycobiota of the investigated soils compared to plants and water-related ecosystems are characterized by a higher figure of number and species composition, but it is not confirmed in the toxigenic mycobiota. So, 56,2% mycobiota of the registered on soils, 68,1% registered on plants and 35,3% registered in the water are contained to the toxigenic fungi.

The result of the analysis of samples taken from investigated cenoses, taken from the materials used for medical purposes, feed and food, have been shown that the dominant species of toxigenic mycobiota includes fungi such as *Aspergillus flavus*, *A.ochraeus*, *Cladosporium herbarium*, *Fusarium moniliforma*, *F.oxysporum*, *Penicillium citrinum* və *P.cyclopium* and their frequencies of the prevalence are between 45,5-57,2% in the area of Azerbaijan.

It was determined that, technogenic impacts cause the change in reduction of species various of mycomplese and eco-trophic specialization, inherent to the Land. So, the formation changes on the base of technogenic impacts that are influenced and characterized by the increase special amount of opportunities and allergens on the fungi biotas which were affected by technological influence of each cenosis and all occasions, and the background level of texnogens are increasing on the relative clean lands according to technological impacts.

The research revealed that the number of fungi in the mycobiota of the plant materials used for different purposes by population, mainly on the medicinal plants, decreases antimicrobial activity. It can be used as a mycology ensure principles of plant materials which used for different purposes by population and can be used as the basic data for the preparation of normative documents.

Bektemirova R.M., Khimich S.D., Kovalchuk V.P., Kondratyuk V.N., Zaitseva T.A., Dyachok Y.R.

A NEW EXPERIMENTAL INFECTED CUTANEOUS WOUND MODEL IN RABBITS

National Pirogov Memorial Medical University, Vinnytsya, Ukraine
rbek.dis@gmail.com

Motivation: Incisional infectious process takes place in about 2/3 of all cases of the surgical site infection. The most common representatives of the hospital-associated microflora are: *Staphylococcus aureus*-20% of all cases; *Escherichia coli* and *Pseudomonas aeruginosa* - 8% and 8%. Wound contaminated with *P.aeruginosa* is quite difficult to treat. Besides, the wound microflora is usually represented by the mixed infection.

Problem statement: During our experiments we faced the problem of receiving the necessary septic skin wounds in the rabbits with 100% reproduction of the certain microflora. The purpose of our study was to develop a reliable model with the maximal reproduction of a predetermined bacterial flora.

Approach: For the study we took 13 mature male and female healthy rabbits. Each animal was in the same conditions: in separate cages and didn't contact with others. Previously they were held 72 h in quarantine.

Approximately 2 cm circular incision was made in the midline between the scapulae through all skin layers. As the rabbit's skin is rather mobile, the wound was fixed by 4 paravertebral sutures. A gauze swab with a 10% solution of calcium chloride was inserted into the wound. After 24 h it was removed. The wound was irrigated with a liquor of decamethoxinum and normal saline to remove foreign flora and antiseptic leftovers. We closed it with a "Tegaderm" membrane, sized 10x15 cm. Experimental infection was produced by a liquid bacterial challenge – an injection of 1 ml of a bacterial suspension directly into the wound (1×10^9 CFU per 1 ml). After 72 h we opened wounds and carried out their photo fixation and analysis of the bacterial inoculum.

Results: The predetermined microflora was obtained in all cases.

Visually, clear signs of septic inflammation in the area of lesion and sutures were detected.

Bacteriology analysis showed that in all cases both microbes were received. An average concentration of *S.aureus* and *P.aeruginosa* was 10^{7-9} CFU.

Conclusions. The developed model offers the high-chance possibility to obtain a wound with a predetermined microflora. It can be used in studies of the septic wounds' treatment.

Berehova Kh.A., Pirog T.P.

ANTIMICROBIAL AND ANTIADHESIVE ACTION OF *NOCARDIA VACCINII* IMV B-7405 SURFACE-ACTIVE SUBSTANCES

National University of Food Technologies, Kiev, Ukraine

khrysty91@ukr.net

Production of surfactants is highly promising direction in the industry, since such connections have several advantages over synthetic analogs. Surfactants are widely used in various industries (environmental technologies, food processing, agriculture, medicine).

The oil-oxidizing bacteria were isolated from the oil-polluted samples of soil and identified as *Nocardia vacinii* IMV B-7405. The ability of these strain to synthesize the metabolites with surface-active and emulsifying properties was determined. It was also determined that the chemical composition of surfactants synthesized by *N. vacinii* IMV B-7405 was a complex of neutral, glyco- and aminolipids. Glycolipids represented by trehalosediacylates and trehalosemycolates.

The aim of this work was to study antimicrobial properties of *N. vacinii* IMV B-7405 surfactants and to study the role of these metabolites in adhesive some microorganisms on the surfaces.

We have shown that *N.vaccinii* IMB B-7405 surfactant can be used for development of effective preparations decreasing the adhesion of microorganisms on the surface. Thus, treatment with the surfactants from strain IMB B-7405 (0.005–0.05 mg/ml) various surfaces (catheters, dentures, plastic, polyvinyl chloride, tiles, and steel) was accompanied by decrease adhesion of bacteria, yeast and micromycetes by 20–85 %. We also studied the antimicrobial properties of biosurfactants. It is shown that the antimicrobial activity of *N. vacinii* IMV B-7405 surfactant depended on the degree of purification (supernatant, solution of surfactant), concentration and exposure. Survival of *Escherichia coli* IEM-1 and *Bacillus subtilis* BT-2 (both vegetative cells and spores) after treatment

for 1–2 hours with surfactants solution and the supernatant (the surfactant concentration 21 mg/ml) was 3–28%. Minimum inhibitory concentrations of *N.vaccinii* IMV B-7405 surfactants on studied bacteria, yeast and micromycetes were 11.5–85.0; 11.5–22.5 and 165.0–325.0 µg/ml respectively. Minimum inhibitory concentrations of *N.vaccinii* IMV B-7405 surfactants are comparable to those of the known microbial surfactants.

Bilkei M., Krivtsova M.

FACULTATIVE MICROBIOTA OF THE RIVER UZH AS AN INDICATOR OF
STRUCTURAL ALTERATIONS OF THE HYDRO ECOSYSTEM IN
ANTHROPOGENIC CONDITIONS

Uzhgorod National University, Uzhgorod

mariannabilkei@ukr.net

The continuous deterioration of surface water quality determines the increasing of monitoring requirements, which particularly affects transboundary watercourses, such as the river Uzh.

The following areas were selected to researches: urbanized which is covers the territory of the city of Uzhgorod; technogenically transformed, situated within the city of Perechyn, where the wood-chemical industrial complex is located; agricultural which is situated in the village Storozhnytsia and recreational. As a result of research it was isolated bacterial strains of the family *Enterobacteriaceae* also their levels and spectra of resistance to antibiotics have been investigated. On the boundaries of the urban area, namely, to the city of Uzhgorod such pathogens were found *Klebsiella spp*, *Escherichia spp* and outside the city of Uzhgorod — *Klebsiella spp*, *Escherichia spp*, *Salmonella spp*, *Proteus spp*. In the area of technogenically-transformed territory, to the village Perechyn — *Escherichia spp*. and behind the village — *Escherichia spp*, *Providencia spp*. In particular, 100 m far from the watercourse of Domoradzh were isolate such bacterial species of genes *Proteus spp*, *Citrobacter spp*, *Edwardsiella spp* and *Escherichia spp*. In the lower reaches of the river Uzh to the village of Storozhnytsya were identified pathogens of the genus *Citrobacter spp* and *Salmonella spp* and beyond the village, isolates belonging to the genus prevailed *Salmonella spp*. Analysis of the antibiotic resistance of isolated strains from surface waters indicated significant resistance to antibiotic drugs. Facultative bacteria of water was the most sensitive to the group of quinolones, namely to norfloxacin, ofloxacin, gatifloxacin, ciprofloxacin and lomefloxacin as well as cefoperazone from the group of beta-lactams. In addition, strains of the genus

Escherichia isolated from natural sources compared with human insulates (sputum) were more resistant to the group of aminoglycosides (amikacin), beta-lactams (imipenem, meropenem) and quinolones (ofloxacin).

The results of research lead to further study of this problem and improve the assessment of the quality of surface waters of the transboundary river Uzh.

Bohdan M.M., Kovalenko O.G., Huliaieva H.B.

WHEAT PLANTS RESISTANCE TO STREAK MOSAIC DISEASE INDUCED BY GLYCAN-GLYCOLIPID COMPLEXES TREATMENT

D.K. Zabolotny Institute of Microbiology and Virology of the NASU Kyiv, Ukraine
b_mi@ukr.net

For modern phytoimmunology, one of the promising approaches to protecting crops from the harmful effects of viral infections are the development of new alternative methods, namely the induction of natural resistance of plants using elicitors of protective reactions.

Have been research of enhancement the viral resistance of *Triticum aestivum* L. to wheat streak mosaic virus by stimulating the phytoimmune system with applied of artificial elicitors: glycans-glycolipid complexes (GGC). In our investigations, we applied GGC of various composition based on glycans, glycolipids and thiosulfonates, which were included glucan (G) *Ganoderma adspersum* (Schulzer) Donk, ramnolipid (RL) *Pseudomonas* sp. SP-17, and methylthiosulfonate (MTS). From these substances have been formed supramolecular structures like liposomes. The following GGCs were investigated: G+RL; G+MTS.

Have been shown changing the structural-functional status of the photosynthetic apparatus (PhA) with being used method of the chlorophyll a fluorescence induction at impact of factors investigating. Was determined the values of the maximum quantum efficiency of PSII (F_v/F_m) and K_{pl} (characterize of effectivity the electron transport chain). Have been detected reduction of F_v/F_m value at increase the K_{pl} value in infected of plants, this shows reduction of efficiency of photo-physical processes in the PhA as well as initiation of degradation of photochemical active PSII complexes. At the same time, seed treatment of GGC contributed to an increase in the F_v/F_m value to almost the level of intact plants with a decrease in the value of K_{pl} , indicating the structural and functional stabilization of the PhA under the action of treatment. Similarly, had changed content of chlorophylls (a+b) in leaves. It was shown that treatment of GGC seeds at a concentration of 100 mg/l contributed to the improvement of the

yield attributes, in particular, the number of ears increased, the number and weight of grains/ear head, 1000 grain weight, which leads to growth of grain yield of wheat in 0.6 times on compared to infected plants.

Borzova N.V., Gudzenko O.V.

**GLYCOSIDASE ACTIVITY OF MICROORGANISMS ISOLATED FROM
VARIOUS SOURCES**

Zabolotny Institute of Microbiology and Virology of National Academy of Sciences of Ukraine, Kyiv, Ukraine

nv_borzova@bigmir.net

The aim of research was to investigate the prevalence of alpha-rhamnosidase and complex mannan-degrading enzymes among microorganisms isolated from various sources. As a result of screening of 448 micromycetes, actinobacteria, bacteria and yeast strains, the active producers of extracellular beta-mannanase and alpha-rhamnosidase were identified. We detected strains of *Penicillium aculeatum*, *P. tardum*, and *P. rugulosum*, which produce a complex of beta-mannanases and alpha-galactosidase activity. Also, strains of thermophilic micromycetes species *Corynascus sepedonium*, *Scytalidium thermophilum* and *Rhizomucor tauricus* with high mannanase activity in culture liquid were detected (10 - 130 U/ml). Two strains of *P. aculeatum* and *P. tardum* showed beta-mannanase, beta-mannosidase and alpha-galactosidase activity. Actinobacteria was shown high potential, 70% of all tested strains showed beta-mannanase activity, their activity ranged from 2 to 55 U/ml. The most active yeast biosynthetic species with mannanase activity were *Cryptococcus albidus*, *C. gastricus*, *C. magnus*, *C. terreus*, *C. laurentii*, *Saccharomyces cerevisiae*, *Williopsis californica*, *Metschnikowia pulcherrima*, *Pichia anomala* and *P. guilliermondii*. The mannan-degrading activity in culture fluid supernatant was ranged from 0.2 to 75 U/ml. Alpha-galactosidase activity was found in two yeast strains (*Debaryomyces polymorphus* and *Debaryomyces hansenii* var. *fabryi*). The high beta-mannanase activity of antarctic strains of *C. victoriae* and *C. terricola* and the prospect of their use as producers of mannan-degrading enzymes are shown for the first time. As a result of screening to ability synthesize alpha-rhamnosidase it was shown that *P. tardum*, *P. rugulosum*, *P. restrictum*, *P. aculeatum*, *C. victoriae* and *C. terricola* strains demonstrated activity which ranged from 0.07 to 0.53 OD/mg of protein. It is shown that the soil and sources of plant origin are the optimal media for the isolation of the enzymes of mannan-degrading complex active producers.

Butsenko L., Pasichyk L., Patyka V.

**SDS-PAAG PROTEIN PROFILES OF *PSEUDOMONAS SYRINGAE* STRAINS,
ISOLATED FROM SEGETAL PLANTS IN WHEAT AGROPHYTOCENOSIS**

Zabolotny Institute of Microbiology and Virology NAS of Ukraine, Academic Zabolotny Str., 154, 03143, Kyiv, Ukraine

plant_path@ukr.net

Modern identification of bacteria is based on multisystem approach and assumes studying of various signs of a microorganism for the purpose of establishment of his taxonomical status. One of methods which allows to carry out identification of microorganisms with high reliability is studying of proteinaceous profiles of the whole cells of bacteria by means of SDS-PAAG electrophoresis. This method allows to identify microorganisms with high precision on species and subspecies levels (pathovar, subspecies, serovar, etc.).

The SDS-PAAG bacterial protein electrophoresis method was used to establish the taxonomic position of the *Pseudomonas syringae* strains, which were isolated from the segetal plants of wheat field. By morphological, biochemical and serological properties these strains were attributed to the phytopathogenic species *P. syringae*. However, the question of the belonging of the *P. syringae* strains isolated from weeds to a specific pathovar remained unclear.

Examined strains have identical SDS-PAAG profiles of whole-cell proteins and contained proteins with a molecular weight of 98 to 17 kDa. Dominating proteins were proteins with a molecular weight of 45 kDa and 30 kDa. In addition to their identity, these SDS-PAAG protein profiles were identical to the protein profiles of the typical strain *P. syringae* pv. *atrofaciens* UKM B-1011T and strains of this pathogen isolated from the diseased wheat plants in Ukraine. Based on a preliminary study of the bacterial properties and the study of SDS-PAAG protein profiles, strains isolated by us from the segetal plants of the wheat field are attributed to *P. syringae* pv. *atrofaciens*.

Thus, from bacteriosis infected and externally healthy weeds in wheat fields, we isolated the causative agent of basal bacteriosis of wheat. The obtained data confirm the status of segetal vegetation as a reservation of bacterial pathogens of cultivated plants.

Chaikovska L.A.¹, Klyuchenko V.V.²**INFLUENCE OF MICROBIAL PREPARATIONS ON THE GRAINS QUALITY OF WINTER WHEAT**¹Crimean Research Institute of Agriculture, Simferopol, Crimea;²Agroindustrial College of the Crimean University, Simferopol, Crimea*ludachaika@mail.ru*

The widespread introduction of biological factors in order to intensify crop production has not only environmental but also economic priority. Because no doubt the feasibility of using microbial drugs, in particular on the basis of phosphate-mobilizing bacteria for optimization of plant nutrition, increasing their productivity and improving grain quality. Among the indicators of the quality of winter wheat grain are particularly important protein, gluten and amino acids. The aim of our study was to determine the effects of pre-sowing bacterization (microbial preparations Polymyxobacterin and Albobacterin) of seeds of winter wheat on grain quality (contents of protein, gluten and aminoacids) in the soil-climatic conditions of the Crimea. Field experiments were carried out on the experimental field of the Crimean agroindustrial College on the backgrounds: 1 – without fertilization, 2 – Amophos (P_{30}), 3 – Amophos (P_{60}), 4 – Amophos (P_{90}); soil – southern chernozem carbonate heavy-loamy. The analysis of the obtained results showed, that the use of microbial preparations for presowing bacterization of winter wheat seeds contributed to the improvement of grain quality (increase in protein and gluten contents). It is established that the most optimal dose of mineral fertilizers, providing a positive effect of bacterization, is fertilizer application at the rate of P_{30} . The protein content in the grain under these conditions increased to 13.2 - 13.8 % against 9.9 % in controls, and gluten – to 30-31.7 % (19.2% in the control). It was showed, that the most effective impact on the content of this indicators in grain took Polymyxobacterin: it provided them an increase in mineral fertilizers and on plots without fertilizer. It is established that the joint use of presowing bacterization and P_{30} increasing aminoacids content in grain: total – 66.9 % against control; essential – 39.4-41.5 % and non essential – 1.6-1.8 times compared with control, which indicates an increase of product quality. Thus, the positive influence of microbial preparations (Polymyxobacterin and Albobacterin) on grain quality of winter wheat in the soil-climatic conditions of the Crimea has been established.

Chuienko A. I., Abdulina D. R., Pysmenna Yu.B.

EXTRACELLULAR LIPASE AND CATALASE ACTIVITY OF MICROSCOPIC FUNGI AND BACTERIA DURING CULTIVATION WITH WASTES OF RUBBER AND POLYMER MATERIALS

Zabolotny Institute of microbiology and virology NAS of Ukraine

helmhammer@ukr.net

Annually, 200000 tons rubber and polymers wastes comes to Ukraine's landfills. Alternative way of their disposal is biodegradation by enzymatic decomposition. The main components of wastes — hydrocarbons can decompose under the influence of catalase, other components are sensitive to the action of lipases.

The aim of the work was to evaluate the enzymatic activity of microscopic fungi and bacteria and their potential for biodegradation of rubber and polymer wastes.

The objects of the study were cultures of microscopic fungi *Aureobasidium pullulans* F-159, *Cladosporium sphaerospermum* F-2442, sulfate-reducing bacteria *Desulfovibrio* sp. UCM B-11503, *Desulfovibrio desulfuricans* UCM B-11501 and hydrocarbon oxidizing bacteria *Pseudomonas pseudoalcaligenes* 109, *Rhodococcus erythropolis* 102. As materials for the experiment, rubber, foamed polyethylene and ethylenevinylacetate were used. Cultivation of microorganisms was carried out in liquid media with the addition of these materials as a Carbon source and inducers of the formation of enzymes.

It has been shown that microscopic fungi intensified the colonization of the surface of the investigated samples and caused their depigmentation, while the bacteria exhibited in 7-14 days altered the characteristics of the studied materials slightly. It has been established that microscopic fungi under cultivation on a mineral medium with investigated materials showed a higher ability to form extracellular lipases (specific activity 12.76–750 units / mg protein) and catalase (12, 25–83.25 units / mg protein) in comparison with the sulfate-reducing bacteria, forming 17.66–98.94 units / mg of protein and 1.21–3.51 units / mg of lipase and catalase protein, respectively. Similar indices of specific enzymatic activity were found for heterotrophic carbohydrate oxidation bacteria *P. pseudoalcaligenes* 109, *R. erythropolis* 102 specific activity of lipase was 14.8-26.8 units / mg protein and catalase 6.27-14.8 units / mg protein, respectively.

It was revealed that ethylene vinyl acetate and rubber are the most promising materials for biodestruction, and the most active destructor of this samples was microscopic fungus *C. sphaerospermum* F-2.

Dabrowska I.V., Kudrevska A.U., Lysenko J.O.

**MICROMYCETES - DESTRUCTORS OF WALL COVERINGS OF THE
ARCHITECTURE MONUMENTS IN KIEV**

Taras Shevchenko National University of Kyiv, Ukraine.

dabrowska_irena@univ.net.ua

In modern anthropogenic environment, microscopic fungi cause large damage to buildings of different uses. At the same time, the problem of biodefence is closely related to human ecology because destructors of various materials are often conditionally pathogenic microorganisms that can cause human disease. Architecture monuments that are actively used require special attention in the areas of research on biomaterial damage. Therefore, the purpose of the work was to determine the mycobiotes of damage to the interior of the monument in Kiev – the church of St. Nicholas (Hall of Organ and Chamber Music). The objects of research became microscopic fungi isolated from the affected areas with different levels of damage.

The following fungi were most commonly encountered: *Alternaria alternata* (4 from 8 plots), *Ulocladium botrytis*, *Cladosporium cladosporioides*, *Penicillium chrysogenum*, *Eurotium repens* (2 from 8 plots).

The following species were predominant in general: *Aspergillus niger* 10^3 - 10^7 CFU/g, *Alternaria alternate* and *Ulocladium botrytis* 10^3 - 10^6 CFU/g, *Paecilomyces variotii* 10^3 - 10^5 CFU/g, *Cladosporium cladosporioides* and *Penicillium brevicompactum* 10^3 - 10^4 CFU/g, *Aspergillus oryzae* and *Absidia corymbifera* 10^2 - 10^4 CFU/g.

Among these species, many representatives are often found on rocky substrates and buildings, in particular: *Penicillium chrysogenum*, *Penicillium verrucosum* var. *corymbiferum*, *Alternaria alternata*, *Cladosporium cladosporioides*, *Aspergillus niger*. In addition, a number of species have been identified that have not previously been found in such biotopes, in particular *Eurotium repens*, *Stachybotrys* sp. and rare species: *Penicillium simplicissimum*, *Penicillium roseopurpureum*, *Penicillium brevicompactum*, *Aspergillus flavipes*. The presence of new species for this biotope, and rare species, may indicate further adaptations and the acquisition of more aggressive properties in anthropogenic environment.

Dankevych L.A.

**REP-PCR ANALYSIS OF SOME GROUPS OF PHYTOPATHOGENIC
BACTERIA**

D.K. Zabolotny Institute of Microbiology and Virology, NASU, Kyiv, Ukraine

ldankevich@ukr.net

The advent polymerase chain reaction have greatly facilitated genomic analyses of microorganisms, provide enhanced capability to characterize and classify strains, and facilitate research to assess its genetic diversity. The diversity of some groups of microorganisms can be assessed in a relatively efficient manner using genomic fingerprinting methods (REP-, AFLP-, RAPD-PCR). Our choice fell on REP-PCR (repetitive element PCR fingerprinting), since this method according to literature data is often used not only to assess the genetic heterogeneity of phytopathogenic bacterial groups, but also to the polyphasic taxonomy of species.

Our investigation estimated the genetic heterogeneity of the following groups of phytopathogenic bacteria: «*Pseudomonas xanthochlora*» and *Pseudomonas* sp. – phylogenetically related to *Pseudomonas fluorescens* and *Pseudomonas marginalis* species; «*Pseudomonas lupini*» and *Pseudomonas* sp. – phylogenetically similar to *Pseudomonas syringae* and *Pseudomonas savastanoi* species; «*Erwinia horticola*» and *Erwinia* sp. – phylogenetically similar to *Erwinia amylovora* and «*Erwinia toxica*» and *Pectobacterium* sp. – phylogenetically similar to *Pectobacterium carotovorum* species. On the example of these groups of phytopathogenic bacteria, the effectiveness of the REP-PCR assay has been confirmed by the correct identification of the bacteria at the species level, provided that the typical representatives of the taxa are present in the studies. We also found that the heterogeneity of BOX, ERIC and REP-profiles of these groups of phytopathogenic bacteria is related to the host plant and climatic and geographical conditions, which is confirmed with literature data. According to some researchers, one of the important reasons for this phenomenon is that the selection of affected plants from one climatic-geographical zone may have an effect on the dispersion of short sequences that are repeated (BOX, REP and ERIC) in the genome of isolated bacteria. This fact requires a more detailed and large-scale study and may be useful both in the study of the dynamics of the pathogen's populations and in the case of its spreading.

Divinsky D.M., Zhornyak O.I.

**INVESTIGATION OF ANTISTAPHYLOCOCCAL ACTIVITY OF NEW
ANTISEPTICS**

National Pirogov Memorial Medical University, Vinnytsia, Ukraine

ndivin@ukr.net

The main negative side of antibiotic therapy is the acquisition of microbial resistance to antibiotics. Particularly important were antibiotic-resistant staphylococcal strains. Antibiotic resistant strains of staphylococci cause qualitatively new types of infectious processes, change the clinical picture of diseases.

The most rational way to solve urgent problems is to search for new antibacterial drugs. Some researchers believe that in the treatment of local infections, priority must be given antiseptics. Such antimicrobial drugs should include domestic drugs decamethoxin, myramistin, decanins, quinolines, which are characterized by a broad antimicrobial spectrum and low toxicity.

The aim: Investigation of antistaphylococcal activity of Nitrones, Decamethoxin, unguent Palisept with decamethoxin.

Materials and methods: Range of three new synthetic compounds Nitron N-arilhinolilazometin was investigated using the method of serial dilutions in different nutrient media for the museum and isolated from patients strains of staphylococci.

Results: The studied preparations Nitrons - 1, 2, 3, Decamethoxin, Palisept showed action against 62 museum and strains isolated from patients cultures of microorganisms. The most high antimicrobial action showed the Nitron 1, Decamethoxin on the staphylococci (0,12-31,25 mg/ml). The minimum bactericidal concentration of unguent Palisept with decamethoxin for antibiotic resistant staphylococci was in the range from 0,48 µg / ml (8,5% of strains) to 31,25 µg / ml (one strain). The bactericidal effect for 84,8% of strains was in the range from 0,97 µg / ml to 7,8 µg / ml of this antiseptic.

Conclusions: Highly effective drugs in vitro are Nitron 1, Decamethoxin, Palisept. Highly sensitive to these drugs remain museum and isolated from patients strains of staphylococci.

In the long run, the data obtained allow us to conclude that the antimicrobial activity of the Nitron 1, the drug Decamethoxin deserve further comprehensive study.

Dvorak K. P., Vorobey N. A., Pukhtaevich P. P.

BACTERIAL DISEASES OF SUGAR BEET

Institute of Plant Physiology and Genetics of the NASU, Kyiv, Ukraine

ekaterina-dvorak@rambler.ru

In relation to the intensification of growing crops technologies and application of new pesticides, innovation systems of fertilization and soil treatment on the background of agro-climatic changes, the issues of phytosanitary control in agroecosystems become actual. Primarily, it concerns the phytopathological condition of agrophytosystems, namely the problems of determination of plant diseases etiology and selection of crop protection products. If earlier agrarians had to minimize the harm of fungal infections in the fields, so today diseases of bacterial origin become more common.

The symptoms of manifestation of bacterial diseases of leaf apparatus and crop roots of sugar beet are described. Cancer root of sugar beet is widespread in many growing regions. The disease preferably appears in August and manifests as growths on the roots that grow gradually and reach a considerable sizes. This outgrowth, is mostly connected with the root by a narrow isthmus and is easily broken.

Tuberculosis tumors have irregular shape, rough surface, and on the top of root are formed most often. Internal tissues of tumors are tight, bright, and are filled with light-yellow rot.

Stripe disease of sugar beet caused by the bacteria *Pseudomonas syringae* pv. *aptata* and manifests as dark-brown, sometimes almost black bars along the petioles, and more often on the main or lateral veins of the leaf.

We isolated *Pseudomonas syringae* strains, which initiate the infectious process once they are on the leaves of sugar beet, leading to appearance of dark necrotic spots. This has been experimentally confirmed by artificial infection of young plants. We demonstrated that the lipids of bacteria cells, isolated from infected leaves of sugar beet, include fatty acids, which are typical for *P. syringae* according to quantitative and qualitative composition.

On the basis of morphological, cultural and biochemical properties in the composition of the microbiota of sugar beet roots with typical symptoms of infestation by tail rot it was determined the presence of *Pantoea agglomerans*, *Pectobacterium carotovorum* and bacteria of the genus *Pseudomonas* and *Bacillus*.

Dzhoraieva S.K., Ivantsova E.K., Sobol N.V., Babuta A.R., Kovalik A.I., Pugacheva O.V.

PECULIARITIES OF MICROBIOCENOSIS OF THE VAGINAL BIOTOPE IN WOMEN WITH UROGENITAL INFECTION CAUSED BY THE OPPORTUNISTIC BIOTHA

State Establishment "Institute of Dermatology and Venerology of National Academy of Medical Sciences of Ukraine", Kharkov, Ukraine.

babuta.anastasia@gmail.com

At the same time the frequency of bacterial vaginosis can up to 80% of medical conditions of the female genital sphere. The study included 144 patients aged 18 to 45 years old with inflammatory diseases of the urogenital tract, caused by opportunistic biota. Women were divided into three age groups: the first group (18-25 years old) - 34 women (23.6%); the second group (26-35 years old) - 77 women (53.5%) and the third group (36-45 years old) - 33 women (22.9%). In women aged 18 to 25 years, there were found 56 strains of bacteria: 31 strains (55.4%) of *Staphylococcus spp.*, 13 strains (23.2%) of *Enterobacteriaceae* family and 12 strains (21.4%) to other taxonomic groups of microorganisms (streptococci, micrococci, corynebacterium). In women aged 26 to 35 years, there were isolated 106 strains of bacteria: 58 (54.7%) belonged to *Staphylococcus* genus, 25 strains (23.6%) to *Enterobacteriaceae* family and 23 strains (21.7%) to other taxonomic groups of microorganisms. In women aged 36 to 45 years, there were detected 46 strains of microorganisms: 60.9% belonged to *Staphylococcus* genus, 28.3% to *Enterobacteriaceae* family and 10.8% of the strains to other taxonomic groups of bacteria. Further, there was determined the susceptibility to antibiotics of the isolated microbial agents. The study reveals that the resistance of staphylococci laboratory strains (85 determinations) to benzylpenicillin was 65.9%, oxacillin - 34.1%, macrolides - 65.9%, lincosamides - 42.3%, aminoglycosides - 41.2 %, tetracyclines - 38.8%, chloramphenicol - 25.9%, quinolones - 23.5%. The study reveals that the resistance of *Enterobacteriaceae* laboratory strains (51 determinations), to ampicillin and amoxiclav - 72.5% and 78.3% accordingly, cephalosporins of the first and second generations - 60.9%, aminoglycosides - 39.2%, nitrofurans - 35.3%, tetracyclines - 31.4%, cephalosporins of the third generation - 29.4%, quinolones - 21.6%, chloramphenicol - 19.6%, monobactams - 11.8%.

The data obtained support the necessity of constant microbiological monitoring of the opportunistic microflora to provide rational treatment of the diseases caused by them.

Dziubliuk N.A., Buligina T.V., Varbanets L.D.

**EFFECT OF *PANTOEA AGGLOMERANS* LIPOPOLISACCHARIDES ON
ACTIVITY OF *BACILLUS* PROTEASES**

D.K. Zabolotny Institute of Microbiology and Virology of the NASU

Nidialkova@gmail.com

The effect of lipopolysaccharides (LPSs) on the protease activity of grampositive and negative bacteria in the world has hardly been studied. *Escherichia coli* OmpT protease is the first example of an enzyme that requires LPS for activity. It was established by Kramer et al. in 2002. And in subsequent years it was shown that Lon protease of *E. coli* is inhibited when interacting with LPS. Since the different microorganisms constantly interact with each other, the influence of their structures on one another can also occur. Therefore, the aim of this work was to study the effect of *Pantoea agglomerans* LPSs, which differ in their composition and some species of biological activity, on the activity of bacillus proteases: *Bacillus thuringiensis* IMV B-7324, *B. thuringiensis* var. *israelensis* IMV B-7465 and *Bacillus* sp. P3. The enzymes were purified on neutral and charged TSK gels Toyopearl HW-55 and DEAE-650 (M) respectively. LPSs were extracted from *P. agglomerans* 7604, 8674, P324, 7969 isolated from rye, cereals, wheat, apple, respectively.

The most active were LPSs of *P. agglomerans* P324 and 8674. LPS of *P. agglomerans* P324 showed narrower spectrum of stimulating activity on protease activity. It increased in 4 times the fibrinolytic activity of protease 1 of *B. thuringiensis* IMB B-7324 and in 3 times the activity of protease 2 of *Bacillus* sp. P3.

The influence of the structural components of *P. agglomerans* P324 LPS on protease activity of bacilli indicate that OPS and lipid A play a significant role in activation of enzymes for fibrin hydrolysis, while core and lipid A - for hydrolysis of elastin. Unlike of the native LPS, isolated structural components inhibited hydrolysis of collagen. It was shown that effect of structural components on the *Bacillus* proteases was strain dependence.

Based on the obtained results, further research will be aimed at finding out of some mechanisms of action of LPS of gram negative bacteria on the protease activity of bacilli. It is connected with the existing interest in identification of the structures of various microorganisms interacting one another.

Filonenko G.V.,¹ Salamanina A.A.,¹ Kyryk D.L.²

CONDITIONALLY-PATHOGENIC MICROBIOTA OF DIFFERENT HABITATS IN
INFANTS WITH CONGENITAL HEART DISEASE

¹ Ukrainian Children's Cardiac Center, Kyiv, Ukraine;

²Shupyk National Medical Academy of Postgraduate Education, Kyiv, Ukraine

baklabccc@ukr.net

Objective: to determine the spectrum conditionally-pathogenic microbiota (CPM) in different habitats in infants who underwent surgery at Ukrainian Children's Cardiac Center

Material and methods: during the period from January to December 2017 has conducted in 2180 microbiological studies in 592 patients.

For identification and determination of antibiotic sensitivity of clinically important microorganisms bacteriological analyzer VITEC 2 COMPACT (bioMerieux) was used. The statistical analysis of species, the number of selected microorganisms and antibiotic sensitivity was performed using a computer program WHONET 5.6.

Results: a positive result was obtained samples in 1764 (80.9%). Gram-positive bacteria were seeded is 69.4% (n=1326), Gram-negative – at 23.2% (n=444), fungi of *Candida* – 7.4% (n=142). Among Gram-positive microorganisms leader takes coagulase-negative staphylococci – 47.9% (n=654). *S.aureus* was isolated from different habitats samples in 22.9% (n=312) of cases, followed by a group of streptococci – 12.3% (n=222). Among Gram-negative organisms first place occupied by Non-fermentative – 15.4% (n=333), most of which were *A. baumannii* – 6.8% (n=148). *Enterobacteriaceae* Gram-negative bacteria were in the second place – 15.1% (n=327) of them were *K. pneumoniae* – 9.5% (n=206). *K. oxytoca* was only 0.1% (n=2) of all the selected negative microorganisms. Increased frequency allocation *Enterobacteriaceae* were producers of beta-lactamases and carbapenemase. Opportunistic biota was isolated from sporum culture – 14.6% (n=278), wound – 2.0% (n=39) and other served as clinical material.

Conclusions. In infants with CHD from different biotopes, a wide spectrum of CPM was isolated the most common were representatives of the genera *Staphylococcus*, *Acinetobacter* and *Klebsiella*. Microecological changes in various biotopes are closely interrelated and manifest themselves in the isolation of identical microorganisms.

Fomina N.S., Fomin O.O., Kondratuk V.M., Prokopchuk Z.M.

**SPECIES SPECTRUM OF MICROORGANISMS ISOLATED FROM PATIENTS
WITH A MINE-EXPLOSIVE TRAUMA AT DIFFERENT STAGES OF
EVACUATION**

National Pirogov memorial medical university, Vinnitsya

fomina.vnmu@gmail.com

The spectrum of microorganisms isolated from fire fighting and mines and explosive injuries is constantly changing. The results of bacteriological investigations of fire and mortar explosive lesions of the limbs of the wounded treated at the Military Medical Clinical Center of the Central Region, Vinnitsya (July-November, 2014, February-March 2015) were analyzed. In the species composition of the microflora of wounds on the 1st week after injury, staphylococci (36.8% of cases) prevailed. Gram-negative bacilli were isolated in 21.1% of cases. *B. cereus* was excised in 36.8% of cases, *C. xerosis* - in 5.3%. So, at the 1st week after injury the wounds microflora were presented by environmental contaminants. In order to prevent infectious complications, a combination of ceftriaxone and metrogyl were prescribed.

Data of antibiotic therapy at the advanced stages of evacuation are unreliable. At the final treatment stage (in our study of MMCC CR), the spectrum of antibacterial drugs administered during the third to fourth weeks of injury was represented by carbopenems, fluoroquinolones and beta-lactamic antibiotics of 3-4th generations.

It is clear that changing antibiotics during treatment or replacing them with reserve drugs also affected the change of the microbes species spectrum. Gram negative bacilli (85.7% of cases) were found to be the leading microflora in the second week of treatment, of which 78.6% were nonfermenting bacteria, one quarter of the strains belonged to the genus *Pseudomonas*, the rest - *Acinetobacter*.

Most microorganisms have been resistant to many antibacterial drugs. Among all isolated strains, 11.9% were multiresistente (MDR), and 67.5% were resistant to broad spectrum of antibiotics (XDR).

On the first week after injury 15.8% of the strains taken and isolated from the material, belonged to strains with increased resistance. On the second week, 71.4% of these strains were isolated, on the third week - 96.9%, on the fourth - 70%, on fifth - 54.5% of strains with an extended resistance spectrum were isolated.

Consequently, the greatest number of pathogens with expanded resistance was isolated at the 3rd week of treatment.

Gadimov A.G., Abbasova Z.I., Gani-zade S.I., Zeynalova E.M., Rasulova S.M.
THE SYMBIOTIC CHARACTERISTICS OF LENS CULINARIS MEDIC WHICH IS
GROWN IN THE CHLORIDE-ACID MEDIUM ON CLAYEY-SANDY SOILS, WITH
THE PARTICIPATION OF POTASSIUM HUMATE

Azerbaijan National Academy of Sciences, Institute of Botany, Baku, Azerbaijan
agadimov@mail.ru

One of the ways to speed up using a useless soils is to establish highly productive and stress tolerant systems which can keep their symbiotic efficiency in the agroecological situations. That is why the researches which are directed to organize high productive agrofitocenosis by way of creating effective symbiosis between the beans and rhizobium bacteria is actual. In the research has been found out the role of potassium humate in the formation of lentil plant (*Lens Culinaris Medic.*) symbiotic system in 0,7% NaCl in the clayey-sandy soils. Lentil seeds are wet with 0.003% potassium humate solution infecting with *Rhizobium leguminosarum* bacteria.

The potassium humate obtained from sapropel occupies a special place among humic preparations, as only in it humic acids content reaches 5%, total nitrogen 2.8%, phosphorus 0.4% and potassium 10%, and also the highest content of microelements; zinc, bor, cobalt, molybdenum, copper.

The sprouts are weakly developed in 0,7% NaCl. Depend on the situation nodules in the root of the plant began to appear 6-9 days after the sprouts were planted and after the 11-15 days legoglobins were formed. The numbers of nodules are changed among 9-44 pieces. In the usual environment potassium humate was fertile condition for growth and forming the nodule. Depending on the active symbiotic potential conditions the nodules changed and formed nearly 36-64,8% of common symbiotic potential and in the budding-flowering period reached its peak point-64,8%. In the usual environment using seeds with potassium humate raised the number of legoglobin nodules from 32,3 to 38,1 at the bloom phase at the ripening phase they are raised from 7,8 to 13,5. But in salty environment they are raised from 16,4 to 21,1 at the flowering phase from 0 to 7 at the ripening phase.

As the receiving results, the softening of the toxic affect of chloride salt to the formation of the lentil plants nodules of potassium humate in clayey-sandy soils stimulated their total and active symbiotic potentials.

Gayda G.Z.¹, Smutok O.V.¹, Klepach H.M.², Stasyuk N.E.¹, Gonchar M.V.¹

MICROBIAL ENZYMES AS THE EFFECTIVE ANALYTICAL TOOLS FOR WINE ANALYSIS

¹Institute of Cell Biology NAS of Ukraine, Lviv, Ukraine;

²Drohobych Ivan Franko State Pedagogical University, Drohobych, Lviv region, Ukraine
galina.gayda@gmail.com

Wine as the final result of a long process of physical, chemical and biological transformations is an inexhaustible subject of research, a source of nutrients and good mood. The positive effect of natural wines on human organism was investigated for many centuries and the latest scientific experiments have demonstrated the beneficial health effects of moderate wine consumption.

To ensure the safety and high quality of the final product it is crucial to control the wine production chain – from the management of soil practices till the analysis of bottled wine. Winemaking therefore requires perfect analytical methods at every stage of the production process. Modern high-throughput approaches require special skills, are time-consuming, expensive and often also have low selectivity. Thus, further development of highly selective and sensitive methods for identifying the key ingredients as well as for monitoring wine contaminants to ensure the safety for human health, is an actual problem.

The aim of our investigations is to develop analytical enzymatic methods, including biosensors, for analysis of basic components of wine (glucose, ethanol, glycerol, L-lactate) and dangerous products (L-Arginine as precursor of carcinogenic ethyl carbamate, as well as ions of heavy and transition metals).

As a result, a lot of analytical approaches were developed. Microbial oxidoreductases (glucose oxidase, alcohol oxidase, glycerol oxidase, glycerol dehydrogenase, flavocytochrome b₂), obtained from the microbial over-producing cells, including recombinant, were used for the analysis of basic components. To elaborate the methods for L-Arginine (Arg) assay, arginase and arginine deiminase, were utilized. The methods of manganese (II) ions assay, based on apoenzyme of arginase, were suggested. The proposed enzymatic-chemical methods and biosensors were tested on the samples of commercial wines. Simple and low-cost enzymatic approaches can be promising in the future for food quality control for determination of enzymes substrates and cofactors, not only for the industrial giants, but also for the small wineries.

Goma Mohamed Huwiage**HELICOBACTER INFECTION AMONG PREGNANT WOMEN IN KASSER
KHIAR AREA PREVALENCE AND RISK FACTORS**

Higher Institute of Medical Sciences and Technology, Algaraboulli, Libya

gomanagoma96@yahoo.com

In 1982 Australian doctors Barry Marshall and Robin Warren at microbiology laboratory at the Royal Australian Hospital in Perth found that *Helicobacter pylori* always found in stomach patients with chronic gastritis and gastric ulcer.

At the first time called it *Campylobacter* CLO, which was assigned to genus *Campylobacter* family *Spirilliaceae*, the genus *Helicobacter* classified in additional eight species: *H.fennelliae*, *H.acinaedi*, *H.mustelae*, *H.maridarum*, *H.nemestrinae*, *H.acinonyx*, *H.rappin*, *H.sputorum*, from these types only *H. pylori* has medical importance.

H. pylori are Gram-negative bacteria, do not form capsules, S-shaped or slightly helical shape, about 2,5-3,5 µm in length, a width of about 0,5-1,0µm, they have 4.6 flagella, which are located in one of the poles of the cell, move fast, even in the thick mucus, microaerophilic, growth at 37°C at 25°C and 42°C are not growing.

In biochemical tests *H. pylori* produce the enzymes hydrogenase, urease, alcoholdehydrogenase, oxidase, catalase, musinase, hemolysine and other enzymes, produces a cytotoxin and specific hemagglutinin.

The infection by *H.pylori* occurs through oral-oral transmission, fecal –oral transmission and gastro-oral transmission.

The aim of this study to determine of *H.pylori* infection among pregnant women in Kasser Khiair city.

Materials and method: One hundred eighty seven pregnant women from 24-38 years participated in this study and divided in four groups according to blood group typing ABO system.

Six ml of blood was drawn and then centrifuged and serum sample obtained then tested by ELISA method, the test was performed according to manufacture instructions.

Results: The study showed that, the prevalence of *H.pylori* among participated cases was not high, from 187 pregnant women only 17 women infected with *H.pylori*, from 17 infected women were 5 cases among O blood group, 3 cases among AB blood group, 5 cases among B blood group, and 4 cases among A blood group.

Conclusion: From this study it can be concluded that the prevalence of *H.pylori* not high in study population cases, and no significant difference according to blood group types.

Gumeniuk I.I., Gruzinskii S.J., Brovko I.S., Chabanyuk Ya.V.

FACTORS OF LEGUME-RHIZOBIUM SYMBIOSIS FORMATION

Institute of Agroecology and Environmental Management of NAAS, Kyiv, Ukraine

gumenyuk.ir@gmail.com

One of the main environmental factors for nodule bacteria is the temperature. It is also known that the optimal conditions for the formation and effective functioning of nitrogen-fixing structures is the temperature range from 15 to 30 °C. At the same time, the temperature regime have a direct effect on the processes of symbiotic nitrogen fixation, which consists inhibiting the nodules formation and indirectly: due to changes in bacterial interactions into the root hairs. Influence of temperature on symbiosis is depending on strain-host specificity. It is known that with temperature changes from 18 to 28 °C, the growth of soybean plants increases and the fixation of molecular nitrogen. Factor which limiting the activity of symbiosis of soybean and nodule bacteria is the upper soil acidity. The powerful soybean symbiotic apparatus formed at optimal pH values from 6.6 to 7. On acidic soils, this plants form less root hairs, the adsorbing capacity of the root system decreases, metabolic processes in the plant slow down. In addition, of own studies and literature sources, this soils contain a significant amount of nodule bacteria, but under such conditions they lose their virulence and activity.

The formation of nodules on the roots of soybean plants by inoculation with Rizoaktiv registered on the 15th day after emergence of the seedlings in the phase of the first trigeminal leaf. Six days later leghemoglobin appeared there and provides energy centers with oxygen and promotes the release of energy to fix atmospheric nitrogen. During the period of 67 days the legume-rhizobium system was formed and actively recorded atmospheric nitrogen in an amount of 127 kg N₂/ ha. The results indicate that 10% of nitrogen is fixed in the phase of the first trigeminal leaf, and in the budding phase – 46% of the total amount of fixed nitrogen.

Therefore, the issue of the influence of environmental factors on the process of formation and functioning of soybean-rhizobium symbiosis remains important. We have found that plants and microorganisms that are in symbiotic interaction are exposed to various external factors that cause inhibition of both, the host plant and rhizobia.

Hrytsev O.A.^{1,3}, Skivka L.M.¹, Kuklin A.V.², Shevchenko J.I.³

DEVELOPMENT OF A TAQMAN REAL-TIME PCR ASSAY FOR THE SPECIFIC DETECTION AND QUANTIFICATION OF *SPORISORIUM REILIANUM* (MAIZE HEAD SMUT)

¹Taras Shevchenko National University of Kyiv, Ukraine;

²Institute of Molecular Biology and Genetics of NASU, Kyiv, Ukraine;

³Syngenta LLC, Kyiv, Ukraine

Olehhytsev@gmail.com

In recent years, there is a deterioration of agroecosystems phytosanitary state in Ukraine, caused by the influence of environmental and economic factors. Large sown areas of maize in specialized farms along with short crop rotation led to the accumulation of soil-borne pathogens, among which causal agent of head smut of corn - *Sporisorium reilianum* (Kühn) Langdon & Full - deserves particular attention. Infection with this fungal pathogen is accompanied by the yield loss up to 15-20%, as well as by a significant deterioration of its quality. To prevent the spread of the disease, it's necessary to identify pathogens in a timely manner and take the necessary agro-technical measures.

The aim of the study was to develop real-time PCR assay for the detection and quantification of *Sporisorium reilianum* DNA in plant samples, using specific primers and TaqMan probes.

NCBI database was used to search for nucleotide sequences and to conduct bioinformatics analysis. The design of specific oligonucleotide primers and TaqMan probes was performed using the online software Primer-BLAST and GenBank database. Alignment of nucleotide sequences was performed using the Clustal Omega software. A primer-probe combination was designed according to the *Sporisorium reilianum* SRZ2 chromosome 7 complete DNA sequence (GenBank accession no. FQ311472.1). We used serial 10-fold standard dilution pDNA, that contains fragments DNA of appropriate phytopathogenic fungi for quantitative determination. Primers and TaqMan probes, based on the nucleotide sequence of the alcohol dehydrogenase 1 gene (GenBank accession no. NM_001111939.2), were designed as internal control for monitoring DNA extraction and PCR inhibition. The reaction mixture was made using Maxima Probe qPCR Master Mix (Thermo Fisher Scientific Inc, USA). Amplification and detection are carried out in a Bio-Rad CFX96 Real-Time Detection System (Bio-Rad Laboratories Ltd., USA).

Huliaieva H.B., Tokovenko I.P., Pasichnyk L.A., Bohdan M.M., Patyka V.P.

PHYSIOLOGO-BIOCHEMIC AND STRUCTURAL CHANGES IN *GALEGA ORIENTALIS* PLANTS AT ARTIFICIAL INOCULATION BY PHYTOPATHOGENIC MICROORGANISMS ISOLATED FROM HOST-PLANTS VARIOUS

D.K. Zabolotny Institute of Microbiology and Virology of the NASU Kyiv, Ukraine
agulaeva34@gmail.com

According to monitoring data, among phytopathogenic microorganisms such as phytopathogenic bacteria and phytoplasma occupy a significant place, in particular, due to a wide range of ecological niches of survival. However, the issue of phylogenetic specialization of certain types of microorganisms remains insufficiently researched. Therefore, the goal of research was to investigate the physico-biochemical and structural changes in plants of *Galega orientalis* at artificial inoculation with strains of phytopathogenic microorganisms isolated from specific plant species.

The decrease in the maximum quantum efficiency of PSII were showed, indicating the destruction of pigment-protein complexes at inoculation by strains of pathogens – *Acholeplasma laidlawii* 118 (specific of wheat) and – 101 and 178 (specific of tomatoes) and mixed inoculation of *A.laidlawii* 118 and *Pseudomonas syringae* pv. *atropfaciens* D13 (specific of wheat).

It was established that at monoinoculation by *A. laidlawii* 118 that are specific to wheat, and especially – at mixed inoculation *A. laidlawii* 118 and *P. syringae* pv. *atropfaciens* D13 there was an increase in the value of K_i (reflects the activity of the RuBPCO). This may be connected with partially with of an increase to oxygenase activity of the enzyme and the rising the proportion of respiration as respondent to stress and substrate-enzyme regulation of photosynthesis evoked of intervention of pathogen in plant metabolism and used plant assimilates by pathogens as source of nutritional, in particular carbon and nitrogen.

Significant decrease in the area of the root system and number of nodules on the roots, and hence reduction the nitrogen-fixing ability of *G. orientalis*, were have been most pronounced in inoculation with strains specific to wheat – monoinoculation *A. laidlawii* 118 and mixed inoculation – *A.laidlawii* 118 and *P. syringae* pv.*atriafaciens* D13.

Thus, the obtained data testify to the potential ability of used in our investigates microorganisms to a wider phylogenetic specialization.

Specific primer pairs and *TaqMan* probes for detection and quantification *Sporisorium reilianum* were developed. The conditions of amplification were established for the real-time PCR for the pathogen DNA detection.

Isayeva V.K., Babayeva I.X., Qasimova S.Y., Aliyeva L.A.

**ACTIVITY OF PEROXIDASE AND POLIPHENOLOXIDASE ENZYMES OF
SOME MICROMYCETES AND SOILS OF AZERBAIJAN**

Institute of Microbiology of Azerbaijan National Academy of Sciences, Baku, Azerbaijan
babayevairada@mail.ru

One of the most important national economic tasks is the improvement of soils and the increase of their fertility. Between the size of the crop yield and the parameters of biological activity of soil, such as nitrogen fixation, cellulolytic activity, enzyme activity, etc., a close correlation is established. At present, the determination of the number of microorganisms in the soil is not so much important as the knowledge of the basic biochemical processes occurring with the participation of microorganisms in carrying out certain agricultural techniques.

It is known that the enzymes of the oxidation-reduction processes play an important role in the soil formation and fertility, as well as the reactions of the process of peroxidase are great importance. However, biochemical studies on the issue in recent years, attention has been paid to the study of peroxidase and polyphenoloxidase enzymes. The purpose of the presented work is study of the peroxidase and polyphenoloxidase activities of the soils in North-East parts of Azerbaijan and the peroxidase activity of micromycetes isolated from these soil samples. As an object of study samples were used 8 soil types in the North East part of Azerbaijan. Determination of soil peroxidase and polyphenoloxidase has been carried out by the method F.X. Xaziyev.

Peroxidase and polyphenoloxidase activity was investigated on the 8 types of soils (sandy, gray-brown, gray, light brown, gray dark-brown, chestnut and dark chestnut, brown-meadow, meadow-forest) of the North East part in the winter and summer seasons. The humus content in soil samples ranged from 0.5% in sandy soil to 3.8% in meadow-forest soil. It was revealed that during the experiment, increasing the amount of humus in the soil decreases the activity of the enzyme peroxidase, but is increased the polyphenoloxidase activity. At value of humus 0.5% in the soil in summer season the maximum peroxidase activity has been - 3.5 mg of purpurogallin. At value of humus 3.8% of meadow-forest soils the peroxidase activity decreases to 2,0 mg of purpurogallin, while polyphenoloxidase activity increases. Thus, it became clear that the peroxidase activity reduces at

increasing amount of humus and productivity of the soil. The results obtained by us are confirmed in similar works by other researchers. Our results indicate that the activity of peroxidase and polyphenoloxidase is inversely proportional. It was clear that the activity of peroxidase and polyphenoloxidase determine the direction of the process of humus becoming. Similar studies conducted on winter soil samples. It was shown that the amount of humus in the soil samples of the summer season is more, the activity of peroxidase and polyphenoloxidase is high. Plants vegetation activity in summer creates favorable conditions for the growth of microorganisms. In the aerobic favorable conditions is on humification of plants' remains and increase polyphenoloxidase activity.

Along with the study of soil enzymes, 150 strains belonging to the genera *Absidia*, *Rhizopus*, *Mucor*, *Geotrichum*, *Aspergillus*, *Paecilomyces*, *Spicaria*, *Penicillium*, *Gliocladium*, *Fusarium*, *Alternaria*, *Stenphulium*, *Cladosporium* were isolated from the plant residues, tree roots and investigated 8 soil types of Azerbaijan. Using the method of color reactions, primary screening was carried out among the isolated strains of fungi. As shown by screening, the greatest number of active strains were isolated from the plant residues. Active strains belong to the genera *Alternaria*, *Cladosporium*, *Geotrichum* and *Penicillium*.

Ivakhnyuk T.V.¹, Molozhavaya O.S.², Makarenko A.N.³

CHARACTERISTICS OF LACTOBACILLUS SPP. AND BIFIDUMBACTEIUM SPP. ISOLATED FROM PATIENTS WITH ALZHEIMER DISEASE

¹Sumy State University;

²Taras Shevchenko National University of Kyiv;

³Interregional Academy of Personnel Management

tivakhnjuk@gmail.com

To date, the study of the relationship between the intestines and the brain, the so-called gut-brain axis, through which the brain carries a modulating effect on the function of the digestive tract, and the latter - on the contrary, regulates the permeability of certain substances through the mucous membrane of the intestine is relevant.

The aim of the work was to study the adhesive properties of gut indigenous microbiota of patients with Alzheimer's disease (AD). The intestinal microflora of 16 patients with AD was studied. The cellular substrate for the study of adhesive activity were formalized human erythrocytes of 0 (I) Rh (+) group.

When analyzing the results of microbiological examination of feces from patients with AD, it was found that in 37.4% of cases registered dysbiosis of the

first degree; in 31.3% - the second degree and 31.3% - the third degree. The amount of *Lactobacillus spp.* and *Bifidobacterium spp.* was significantly decreased ($p < 0,05$) compared with the control group - elderly people of $75 \pm 0,9$ years old. The increased levels of *Lactobacillus spp.* ($lg\ 4.48 \pm 0.15$ CFU / g) was registered in patients with AD with a third degree of dysbiosis, and decreased levels of *Bifidobacterium spp.* ($lg\ 3.7 \pm 0.2$ CFU / g) - in patients with II degree of dysbiosis -

Results of the study of adhesive activity of strains *Bifidobacterium spp.* and *Lactobacillus spp.*, isolated from patients with AD showed that 83.3% and 60.0% of *Lactobacillus spp.* isolated from patients with I and II degree of dysbiosis showed a high and average adhesive activity; 80% of *Lactobacillus spp.* isolated from patients with the third degree of dysbiosis showed low adhesive activity (1.72 ± 0.08).

When cultivating *Bifidobacterium spp.* and *Lactobacillus spp.* in hydrolyzed milk as a monoculture (pH 7.2; 37° C), the adhesive properties of low-adhesion strains was significantly increased ($p < 0.05$) compared with the previous results.

These studies of the antagonistic activity showed the possibility of using *Bifidobacterium spp.* and *Lactobacillus spp.* strains in the form of an autobiotic and is a good way to develop a personalized drug for correction of gut dysbiosis in patients with AD.

Ivanova S., Kryzhanovskaya A., Martynchuk A.

THE PROBIOTICS AND THEIR SIGNIFICANCE

National Pirogov Memorial Medical University, Vinnytsya, Ukraine

2205avk1965@gmail.com

Microflora of the human body is involved in numerous vital bodily functions. An important role is given to autochthonous microflora in the digestion and utilization of nutrient substrates, regulation of water-electrolyte and gas energy exchange, heat exchange, synthesis of vitamins and biologically active compounds (bacteriocin, lysozyme, antibiotic-like substances, cytokines), etc. Maintenance of the microbiocenosis of the organism is achieved through the interaction of immune and digestive systems. Therefore, the disturbance of their functions leads to the development of dysbiosis. Correction of microbiocenosis disturbance is possible only with simultaneous action in two directions. First, is the elimination of the underlying disease, which led to dysbiosis, levelling secretory, motor-evacuation disorders. Second, is the restoration of the native microflora of the intestine. The use of probiotics in the treatment of dysbiosis in a practice is

complicated by a number of circumstances. Probiotics are not sufficiently resistant to fluctuations in pH, osmotic pressure, bile and enzyme activity, chemotherapy and radiation therapy. It is difficult to pick up biopreparations for a particular patient, since they are phenotypically and genotypically alien to this individual and, therefore, are poorly curable. The mechanism of curative action of probiotics is not only in the simple colonization of the intestine. First, it is the inhibition of allochthonic microflora (colonization resistance) adhesion, competition for nutrients, products of antimicrobial substances, acidification of the intestinal contents, stimulation of the immune response (synthesis of secretory Ig), synthesis of cytoprotective substances. Secondly, probiotics containing *Lactobacillus* and *Bifidobacterium*, to a large extent, have immunostimulatory activity. Probiotics with *Escherichia coli* have a more antagonistic effect, and a number of strains transforms cholesterol.

Ivashchenko O.Y.¹, Livinska O.P.², , Kovalenko N.K.²

**THE INFLUENCE OF HYDROGEN PEROXIDE PREADAPTATION ON
BIOLOGICAL PROPERTIES OF *LACTOBACILLUS PLANTARUM* STRAINS**

¹Taras Shevchenko National university of Kyiv, Ukraine;

²D.K. Zabolotny Institute of Microbiology and Virology of the NASU, Kyiv, Ukraine

ivaschenko@i.ua

The oxidative stress and preadaptation effects on lactobacilli strains are currently of great importance and also might provide fundamental as well as applied interest. In this regard, special attention is drawn to the studies that could provide the opportunity to establish the mechanisms of stress resistance increase of lactic acid bacteria (LAB) under industrial conditions. The capacity to adhere to macroorganism epithelium, antagonistic activity against pathogenic microorganisms and acidifying activity are considered to be important properties of industrial probiotic strains of lactobacilli.

The aim of this study was to investigate the effect of preadaptation with hormetic doses of hydrogen peroxide on the adhesive, antagonistic and acidifying activity of *Lactobacillus plantarum* strains.

The objects of the study were the strains *L. plantarum* 11/16, *L. plantarum* 47sm and *L. plantarum* 1/12UAE. Adhesive properties of microorganisms were assessed by determining the medium adhesive index (MAI), the coefficient of involvement of epithelial cells in the adhesion process (K) and the index of microorganisms adhesion (IMA). Antagonistic activity was investigated by the method of delayed antagonism against pathogenic and

opportunistic reference strains. The acidifying activity of lactobacilli was investigated in 0.5% skimmed milk by titrimetric method. The preadaptation of cells was performed by incubating in the presence of hydrogen peroxide in the concentrations that exhibited the hormetic effects for the strain.

It was shown, that adhesive activity of the studied strains was not species- or source- depended but depended on the effect of hydrogen peroxide: adhesive properties were increased due to the exposure of hydrogen peroxide preadaptation by 20-100%. Effective preadaptation doses of hydrogen peroxide mostly inhibited the antagonistic properties of the strains. There was a slight acidity index decrease of the strains *L. plantarum* 11/16 and 47sm after the exposure of effective preadaptation doses of H₂O₂, whereas the strain *L. plantarum* 1/12UAE did not demonstrate such effect.

Ivchuk V.V., Kovalchuk T.A.

THE MICROBIOM OF RESPIRATORY TRACT IN PATIENTS WITH CHRONIC OBSTRUCTIVE DISEASES OF LUNGS OF PROFESSIONAL ETIOLOGY AND ITS SENSITIVITY TO CHEMOTHERAPEUTIC DRUGS

Ukrainian Research Institute of Industrial Medicine, Kryvyi Rih, Ukraine

ivchukv@yahoo.com

One of the main causes of exacerbation of COPD of professional etiology may be microbial infection, which, according to scientific literature, causes about 80% of all exacerbations. The question of the use of antibacterial drugs in exacerbation of COPD of professional etiology remains relevant today.

The purpose of the work was to investigate the antibiotic resistance of strains of microorganisms isolated from sputum in patients with COPD of professional etiology of workers of the mining industry.

The results of our bacteriological studies showed that the dominant microorganisms in the sputum patients with exacerbation of COPD of professional etiology were: *S. aureus* (26.7 %), *C. albicans* (22.3 %), *K. pneumoniae* (10.9 %), *E. coli* (9.3 %), *S. viridans* (5.7 %) and *S. epidermidis* (5.5 %). The highest sensitivity of gram-positive microorganisms culture was to cephalosporin III generation – cefoperazone, from 8.6 % to 12.5 %. According to our data, gram-negative bacteria isolated from patients with COPD of professional etiology showed sensitivity to cephalosporins 1.4 % - 12.6 %, aminoglycosides 1.7 % - 11.5 %, chloramphenicol 9.0 % - 12.0 %, quinolones 1.1 % - 11.5 %. Gram-negative bacteria had the highest sensitivity to cefoperazone, from 5.6 % to 12.6 %. Also, high sensitivity, 5.5 % - 10.6 %, was characteristic of

ceftriaxone. Our studies showed that *C. albicans* were the most susceptible to clotrimazole – 34.1 %. In terms of its antifungal activity, nystatin is in the second place – 25.7 %, after clotrimazole.

The leading positions in the species composition of the respiratory microflora of patients with COPD of occupational etiology include: *S. aureus*, *C. albicans*, *K. pneumoniae*, *E. coli*. The most sensitive isolated bacteria were to the antibiotics of the cephalosporins group. The antimycotic clotrimazole exhibited the highest activity in relation to fungi of *C. albicans* species.

Kachur T.L., Gretskey I.A., Zelena L.B.

**ELECTROMAGNETIC RADIATION AS THE FACTOR OF YEAST
FLOCCULATION INCREASING**

Zabolotny Institute of Microbiology and Virology, NAS of Ukraine
tkachur07@gmail.com

Flocculation is the reversible aggregation or agglutination of yeast cells. This feature of yeast is widely exploited in brewing and winemaking. During the fermentation yeasts consume sugar and at the end of the process they produce flaky structures and drop down. Yeast flocculation is associated with biological and colloidal stability; it affects wort fermentation and organoleptic characteristics. The various approaches and techniques as well as yeast selection are used to increase flocculation. One of the factors that may have biological effects on organisms including some metabolic changes is electromagnetic irradiation.

The aim of the present study was to assess the influence of EMR on yeast flocculation process. Two strains of *Saccharomyces cerevisiae* from Ukrainian Collection of Microorganisms of Zabolotny Institute of Microbiology and Virology were cultured in YEPD broth. Yeast cell suspension was exposed to two types of EMR: extremely high frequency (61,2 GHz and 0,2 mW) for 30, 60, 90 min and ultrahigh frequency (2,45 GHz, 15 W) for 15 min. Evaluation of flocculation ability was carried out with a microflocculation technique.

Results of analysis revealed that yeast cells subjected to EHF EMR showed enlargement of flocculation index up to 15% while the exposure duration was increasing. A very slight increasing of flocculent cells (up to 7%) was observed after UHF EMR treatment for 15 min. Results obtained supposed that EMR exposure can increase the level of yeast flocculation but the irradiation regime should be carefully defined.

Kameneva I.A., Yakubovskaya A.I., Gritchin M.V., Smirnova I.I., Konopleva G.N.

**DEVELOPMENT OF MICROBIOLOGICAL COMPLEX FOR PLANT RESIDUES
BIODESTRUCTION**

Federal State Budget Scientific Institution «Research Institute of Agriculture of Crimea»
irina.kameneva.7@mail.ru

Microorganisms are destructors of organic matter as well as important components of pedocoenosis. Due to their activity, the biogenic elements are cycled and, consequently, they are vital to agriculture through their role in maintaining soil fertility. Development of effective technologies for the use of plant residues is an urgent task of agriculture.

The purpose of the research is to create an effective microbiological complex for the destruction of cereal crops residues.

Associations of aerobic microorganisms with high rate of cellulolytic activity were identified from different ecotopes. Microbiological analysis has shown that non-spore bacteria, bacilli, algal flora, micromycetes and actinomycetes are the constituent components of aforementioned associations. Optimal parameters of deep cultivation of cellulolytic associations have been established. Collection of bacterial strains of different functionality were selected to form a microbiological complex. In vitro research had shown the stability of the microbiological complex to abiotic stress factors (high temperature, moisture deficiency). Treatment of cereal crops residues with the microbiological complex doubles the rate of organic material decomposition. A positive effect of the microbiological complex on the microbiota of soil and rhizosphere of winter wheat has been established. The number of microorganisms, which absorb nitrogen of mineral compounds, rises by 43% in comparison with the control and the intensity of mineralization processes is almost doubled. An increase in soil organic matter was noted. The application of the microbiological complex in agrocenoses of winter wheat contributes to its bio-productivity improvement.

Thus, microbiological complex, which includes collection of high-efficiency bacterial strains of different functionality and association of microorganisms that break down cellulose, has been developed to destruct plant residues.

Karpenko V.V., Oryabinska L.B., Bogdan T.Z.

PREPARATION OF THE CELL LYSATES FROM *LACTOBACILLUS* STRAINS

National Technical University of Ukraine "Igor Sikorsky Kyiv Polytechnic Institute"

vicky.st.cl@gmail.com

In recent years, due to the expanding allergic population, it is noticed an increasing interest to the drugs from living microorganisms – representatives of human normobiocenosis. The great attention of pharmaceutical enterprises is paid to the lyophilized mass of lactic acid bacteria, while the lysates, enriched with cell wall fragments and components of cellular content, have strong immunomodulatory properties. The purpose of the investigation was to find the optimal conditions for the production of lysates of lactic acid bacteria genus *Lactobacillus*.

In this study 12 strains of lactobacilli from the Museum of Cultures of the Department of Industrial Biotechnology FBT, NTUU "Ihor Sikorsky KPI" were used. The lysates were obtained by the ultrasonic disintegration (acoustic power of the recipient 40 W / cm², ampacity 0.3-0.4 A, oscillating frequency 22 kHz) and the enzymatic lysis using enzymes: lysoreciphene from the culture fluid of the strain *S. recifensis subsp. lyticus* 2435 (98.00-100.000 U / ml) and lysozyme (100 and 200 µg / ml). Hydrolysis of cells was carried out in a water bath at a temperature of 40 ° C. and 50 ° C for 30-120 minutes. The concentration of cells in the reaction mixture was 10%. Lytic sensitivity (LS) of bacteria was determined by changing the optical density of cell suspension at a wavelength of 540 nm. The analysis of the results allowed to establish that *L. plantarum* 2621 (LS = 64.12%) and *L. murinus* LE (LS = 43.36%) were the most susceptible to lysorecifenum strains when processing cells for 120 min at temperature 50 °C. The highest sensitivity to lysozyme (200 µg / ml) was shown by *L. delbrueskii subsp bulgaricus* LB51 (LS = 67.8%) and *L. delbrueskii subsp. bulgaricus* LB86 (LD = 57.1%) strains at 55 °C during 90 min. Thus, the optimal way of obtaining of the lactic acid bacteria lysates is enzymatic disintegration, while ultrasonic disintegration was not effective.

Katiukha V.L. , Kovalchuk V.P.

TO THE QUESTION OF EFFECTIVE HYGIENE OF HANDS OF MEDICAL PERSONNEL IN THE PREVENTION OF HOSPITAL INFECTIONS

National Pirogov Memorial Medical University, Vinnytsya

serafim79@i.ua

The problem of hospital infections has become so urgent that the World Health Organization is forced to take coordinated efforts aimed at resolving it. It is

no coincidence that in the WHO guidelines on this issue, much attention is devoted to the means and the proper technique of handling the hands of medical personnel. It has been proved that today most nosocomial infections are transmitted by the hands of medical personnel. This situation, on the one hand, is explained by the non-compliance by the staff with the rules of hygienic hand treatment, but more often - by the inadequate effectiveness of the drugs used for hygiene purposes.

The modern domestic arsenal of means for disinfection of the hands of medical personnel has many trade marks, while it is based mainly on alcohol solutions that do not meet the modern requirements for antiseptics. Such preparations are flammable, do not act on spore-forming bacteria and non-cancerous viruses, violate the integrity of the skin.

One of the antiseptic drugs, which does not have the above-mentioned disadvantages, is the domestic antiseptic decamethoxin from the group of surfactants. The drug has a wide range of antimicrobial activity. The resident coccal skin microflora decamethoxin has a bactericidal effect at a concentration of 1-5 μg / ml. Conditionally pathogenic bacteria from the *Enterobacteriaceae* family, which are frequent pathogens of intracerebral infections, die in concentrations of the drug no more than 15-30 μg / ml. Being a surfactant, the drug exhibits detergent properties, while it does not apply to the skin of drying action, a similar effect of alcohols. It should be recognized that the use of this domestic drug for disinfecting the hands of the medical staff in order to prevent the spread of hospital infections.

Kolomiets J.V.

INFLUENCE OF BACTERIAL PATHOGENS ON THE FATTY ACID COMPOSITION OF TOTAL LIPIDS OF TOMATOES CALLUS

National University of Life and Environmental Sciences of Ukraine, Kyiv, Ukraine

julyja@i.ua

In the plant resistance against bacterial infection in addition to specific, an important role is played by non-specific response of cells at the level of membranes. The effect of bacterial stress on lipid fatty acid (FA) composition of tomato plants is insufficiently studied. The purpose is studying of the FA composition of total lipids of callus tissues of tomato varieties in the conditions of bacterial stress. The objects of research were callus cultures of tomato varieties Oberih and Flora with different resistance to bacteriosis pathogens. In the experiments that simulated the impact of stress factors, to the basic culture medium it was added 4,0 % inactivated cells (IC) strains isolated on the farms of

Ukraine isolates *P. syringae* pv. tomato IZ-9, *X. vesicatoria* IZ-10 and *C. michiganensis* subsp. *michiganensis* IZ-58. Lipids were extracted by a modified Bligh and Dyer's method. Adding into the nutrient medium of IC pathogenic strains caused growth of unsaturated FA which takes place through acyl-lipid desaturase, evaluation of the activity of which were determined through stearoil-(SRD), oleil-(ORD) and linoleil-(LRD) desaturase ratios. For tomato varieties Oberih and Flora SRD level in the control was in the range of 0,84 – 0,87. In terms of the impact of bacterial stress, the SRD index for Oberih variety increased up to 0,95, and for Flora variety – decreased down to 0,64. Activity of acyl-lipid ω 6- and ω 3-chloroplast desaturases were determined through the ratio LRD and ORD, the level of which for callus tissues of Oberih variety was 0,16 and 0,30, and for Flora variety – 0,14 and 0,25. For media with 4 % IC the LRD level for Oberih tomato variety increased when compared to the control and fluctuated in the range of 0,19 – 0,28, and ORD – 0,54 – 0,61. Under these conditions, Flora variety was characterized by increased level of ORD, which was 0,53 – 0,61, and reduction of LRD – 0,04 – 0,09 when compared to the control. It is shown that the response of callus cells of tomato plants to bacterial stress appears in increase of the amount of unsaturated FA by 20,4 – 28,2 %, which is provided by the activation of acyl-lipid ω 6- and ω 3-chloroplastic desaturases.

Kondratiuk V.M.¹ , Kopaneva L.O.²

**MICROBIOLOGICAL CHARACTERISTICS OF THE PRIMARY
CONTAMINATION OF WAR WOUNDS**

¹National Pirogov memorial medical university, Vinnitsya, Ukraine;

²Novoaidarsky RTMO

kondratuk2007@gmail.com

There are a few studies focused on bacteria that contaminate the wound at the time of injury. We cultured the wounds of extremities obtained war in the East of Ukraine in order to investigate the quantitative level of contamination and antibiotic resistance of recovered microorganisms. Bacteriological examination of the wounds during the first hours between the injury and primary surgical debridement was carried out prospectively in the 59 Military Mobile Hospital in 2016. The identification of microorganisms, antibacterial sensitivity, quantity of each species was carried out in the bacteriological laboratory of Novoaidarsky RTMO. Ninety swabs were taken from 84 wounded during 2016. Gunshot wounds accounted for 41% (37 injuries), the rest of the injuries were blunt. Sixty swabs were taken from isolated wounds of soft tissues, 5 were from amputation

sites, the rest were taken from the gunshot fractures. The growth of microorganisms was not detected in 57 swabs (63%). The positive cultures (33 swabs) provided 14 *S. epidermidis* strains, 12 *S. aureus* and 6 *E. coli* strains. Among 14 *S. epidermidis* strains, three of them were resistant to oxacillin, 2 strains were resistant to ciprofloxacin, and all strains were resistant to erythromycin, penicillin and lincomycin. However, 100% of strains remained susceptible to gentamicin. Isolated *S. aureus* strains demonstrated susceptibility to erythromycin, lincomycin, gentamicin, two strains were resistant to vancomycin and ciprofloxacin. However, *S. aureus* strains demonstrated 100% resistance to penicillin and 80% resistance (9 strains) to oxacillin. The isolated *E. coli* strains were resistant to all studied antibiotics except ampicillin / sulbactam, to which all strains were susceptible. The number of isolated microorganisms, regardless of species, did not exceed 103 KFU/g. Detailed data about swab-culture results are available at the scientific depository at: <https://figshare.com/s/a5b195f56a06981d1672>. Unlike previous observations, there are strains that fall under the definition of antibiotic resistant. These data may shape the empiric choice of antimicrobial agents to adequately control contamination.

Kornienko N., Kot T., Kharina A.

ISOLATION OF BACTERIUM *ENTEROBACTER CLOACAE* AND DETECTION OF PHYTOPATHOGENIC PROPERTIES ON ONIONS AND POTATOES

Taras Shevchenko National University of Kyiv, Kyiv, Ukraine

tashe404@gmail.com

Bacteria of genres *Serratia*, *Enterobacter* and *Enterococcus* more frequently happen in the cases of nosocomial infections, e.g. food poisonings and sepsises. But the last researches, however, prove that these human pathogenic bacterial species also have ability to colonize a wide spectrum of plants and cause the disease development. Our purpose was to isolate polybiotrophic bacteria and to investigate phytopathogenic effects they cause to plants.

Vegetables with typical symptoms - brown and black rot of fruits, stalks, were selected in a clean container. Further in laboratory fruits were washed out sterile distillate three times, thin skins over places of rot and necrotic spots processed 72% ethyl alcohol, cut and selected a part of contents for further work. Biochemical and morphological tests were used to identify bacterial species. Among the isolated bacteria there was one which was of the greatest interest as it is the representative of opportunistic flora of the person and it can cause serious

illnesses - *Enterobacter cloacae*. Antimicrobial susceptibility test using the 'Vitek-2' analyzer indicated the resistance of isolated *E. cloacae* to certain antimicrobial substances. The resistance to amoxicillin, cefalexin, a cefpodoxime, an enrofloxacin has been revealed.

Further we checked ability of a bacterium to cause symptoms of bacterioses on potatoes and onions. For this purpose we have taken separate scales of onions and pieces of potatoes (size of 2 cm) and have placed them on sterile filter paper in Petri dishes in which pieces of plants were processed by the night culture of a bacterium (a titre of 10^8). The next days we observed emergence of bacterial decay of brown color on scales of onions and potatoes in the place of processing. On second day samples were half rotten while control remained clean.

It has been shown that the polybiotrophic opportunistic bacterium can cause symptoms of bacterioses on popular vegetable cultures. Taking resistance of this bacterium to antibiotics in attention, we consider it necessary to reconsider methods of processing of such plants and to strengthen control on use of antibiotics in the agrarian industry.

Korobkova K.S.

CURRENT RESEARCHES IN MYCOPLASMOLOGY IN ZABOLOTNY INSTITUTE OF MICROBIOLOGY AND VIROLOGY NASC OF UKRAINE

Zabolotny Institute of microbiology and virology NASc of Ukraine, Kyiv, Ukraine

kkorobkova@ukr.net

Mycoplasmas (*Mollicutes*) are the smallest free-living wall-less procaryotes which are widespread in nature and many are animal, plant and human pathogens. The frequent contamination of cell cultures with mycoplasmas, together with their possession of the smallest genome of any free-living organism, has drawn researcher's interest to these organisms. These microorganisms can cause serious and devastating problems for crop cultures, especially in developing economy countries.

Mycoplasmas are very sensitive to cultivation conditions, the plant pathogenic strains especially. Therefore in the Zabolotny Institute of microbiology and virology it was created the museum of Mollicutes cultures. It is stored in the department of Plant pathogenic bacteria and is a part of the Ukrainian National collection of microbial cultures and counts more than 40 species and strains of mycoplasmas. The founder and for a long time the head of the researching in mycoplasmaology was Corresponding Member of the National Academy of

Sciences of Ukraine Professor I.G. Skripal. At present, researches on plant pathogenic mycoplasmas are continuing under the direction of Academician of the National Academy of Agrarian Sciences, V.P. Patyka.

The physiology of plant yellows diseases is very poorly understood. Simultaneously with the study of the fundamental peculiarities of the mollicutes, our researchers study of the substances influence of different nature in order to control of mycoplasma activity. The relation of mycoplasma invasion to plant phloem dysfunction, hormonal imbalance, toxic metabolites production and, ultimately, symptom expression are the task of our researches. Today our activities are directed at studying the foundations for the realization of plant pathogenic properties of the class *Mollicutes* representatives, the features of the signal and metabolic relations of mollicutes and host macro-organism cells, as well as the changes occurring in the plant organism under the influence of mycoplasma infection.

Kovalchuk V.P.¹, Kishchuk V.V.¹, Vovk I.M.¹, Isnyuk A.S.¹, Kovalenko I.M.¹

ANTIMICROBIAL ACTION OF PLANT AND SYNTHETIC ANTISEPTICS ON BACTERIAL PATHOGENS OF UPPER RESPIRATORY TRACT INFECTIONS

¹National Pirogov memorial medical university, Vinnitsya, Ukraine

darkferor@gmail.com

To make microbiological substantiation and optimize local etiotropic therapy of bacterial infections in the upper respiratory tract we conducted a comparative study of the antimicrobial efficacy of synthetic (decamethoxin, myramystin) and plant (chlorophyllipt, sangvirin) antiseptics on clinical strains of *S.aureus*, *K.rhinoscleromatis*. The antiseptics' antimicrobial action was studied by determination of the minimal cidal concentrations (MCC) with serial dilution test, and compared with results of quantitative suspension test, which was made to find out decontaminating activity of the antiseptics.

As a result of the conducted studies, it was found that *S.aureus* strains had a higher sensitivity to synthetic surface-active antiseptics (MCC was 0.24-3.13 µg/ml) than the strains of *K.rhinoscleromatis*: the corresponding concentrations did not exceed 7.8 µg/ml. But klebsiella strains were more susceptible to sangvirin than staphylococci (MCC was 1.9 µg/ml and 25 µg/ml, respectively). Chlorophyllipt was more effective in staphylococcal strains (the cidal effect was determined at concentration of 62.5 µg/ml, which was 5-10 times less than the corresponding data for klebsiella). According to the results of the suspension test, preparations of synthetic antiseptics (decasan, myramystin) had a rapid

decontamination effect on planktonic forms of both species' experimental strains. Microbial suspensions' total decontamination (1010 CFU/ml) with synthetic antiseptics occurred in 3 minutes. The decontaminative effect of plant antiseptics detected with suspension test was weaker: preparations of chlorophyllipt and sangviritrin at concentrations recommended for irrigation of mucous membranes, reduced the number of staphylococci in 150-300 times, and klebsiella, respectively, 6-10 times in 15 minutes. So, a quantitative suspension test allows us to make more accurate and reliable comparison between antiseptics and evaluate the expected action of local antimicrobials on the mucous membranes. Thus, synthetic surface-active antiseptics had a more pronounced antimicrobial effect and a rapid decontamination effect on studied strains of microorganisms.

Kovalchuk V.P., Prokopchuk Z.M., Burkot V.M.

CHARACTERISTICS OF ADAPTIVE PROPERTIES OF NONFERMENTING BACTERIA

National Pirogov Memorial Medical University, Vinnytsya Ministry of Health of Ukraine, Vinnytsya, Ukraine

burkotvita@gmail.com

The basis of the global problem of antibiotic resistance of bacteria is the general biological mechanisms of living organisms adaptation to changes in the conditions of existence. The emergence inside of the population of individuals surviving at adverse conditions ensures the evolutionary process and conservation of biological species. The speed of the adaptive process of different species varies in a wide range. A group of gram-negative non-fermenting bacteria is characterized by high rates of resistance to antibiotics. The resistance of "wild" strains of *P.aeruginosa* and *A. baumannii* to carbapenems and aminoglycosides antibiotics were studied by the method of successive passages in a liquid nutrient medium with increasing concentrations of antibiotics at artificial conditions. The results of the research have shown that the spin-like increase in the stability of 8-10 times in the representatives of both studied species is already at III-VI passages.

This is can be explained by expression under the influence of antibiotics existing in the studied bacteria strains at the genetic level of adaptive potential. Subsequently, the process significantly slows down and varies in speed at the strain level. By the XV passage stability of acinetobacteria increased in 4-8 times. At the same time, the speed of adaptation did not depend on the initial level of stability.

The same increase the resistance of pseudomonads occurred earlier (on the XIX-X passage) with a pronounced tendency for more rapid adaptation of strains that originally had a higher level of resistance. There are no significant differences in the dynamics of akinetobacteria adaptation to different antibiotics. Pseudomonads quickly formed resistance to aminoglycoside antibiotics and more slowly - to carbapenems.

Thus, the rate of adaptation to life in a medium with high concentrations of antibiotics of the aminoglycoside and carbapenems structure in gram-negative nonfermenting bacteria has differences in the species and strain level. In this case, there is a tendency to preferences in the adaptive potential of pseudomonads, in comparison with akinetobacteria.

Kozlovska G.V.

RESEARCH OF ECOLOGY OF *YERSINIA ENTEROCOLITICA* - IMPORTANT ELEMENT OF DEVELOPMENT OF FOOD SAFETY CONTROL SYSTEM

National University of life and environmental science of Ukraine, Kyiv

annakozlovska @i.ua

Today, the HACCP system (Hazard Analysis and Critical Control Points) is considered to be the most effective system for monitoring the quality and safety of food products. Development of this system require a clear understanding of the biology and ecology of the relevant pathogens, including *Yersinia enterocolitica*.

It should be noted that the most effective prevention of a danger is real in a case of right influence at the pathogen not only at some particular interval, but, if possible, at different stages of its circulation. Circulation cycle in *Yersinia enterocolitica* is extremely complex and multi-vector. Analyzing epizootic (epidemic) process is taken to determine the reservoir, and a source of transmission factors, the susceptible organism, to name the mono-vector migration cycle of the pathogen in nature.

In particular, in the case of *Y. enterocolitica*, the variant "animal-soil-plant-animal" is proposed (Skibitsky VG, 2012). At the same time, the cycle of migration of this microorganism is too complex to be reproduced in mono-vector form.

An analysis of the relevant literature reports and our own studies suggest the following model scheme for the circulation of *Yersinia enterocolitica* in nature. The basic cycle of circulation of *Y. enterocolitica*: "Fauna - abiotic substrates - flora - fauna" (the sequence of phases can be different). "Mini-cycle" of circulation of *Y. enterocolitica*: •Fauna - Flora (source of man or animal, object of infection - plants);

- Flora - Fauna (source - plants, the object of infection - people, animals);
- Fauna - Fauna (source of animals, the object of infection is similar - animals);
- Flora - Flora (the source of the plant, the object of infection is the same - plants).

In each case, various transmission factors of the causative agent (abiotic substrates, mechanical, biological) may appear. The circulation of *Yersinia enterocolitica* occurs both within certain minicycles and between them.

In view of the foregoing, we are developing tools and methods for detecting *Yersinia enterocolitica* in pathological material, food products, animal feed and environmental objects.

Krytsova M.V.¹, Livak O.G.², Balabanska B.V.¹, Ganich T.T., Marochka N.A.¹, Bilak O.M.¹, Kuklak Ch. T.¹, Chicherska M.V.¹, Perestuk A.S.¹
ANTIBACTERIAL ACTIVITY OF ESSENTIAL OILS AGAINST CLINICAL ISOLATES OF FACULTATIVE MICROBIOTA

¹Uzhhorod National University, Uzhhorod, Ukraine;

²Svaliava Central District Hospital, Svalava, Ukraine

maryna.krivcova@gmail.com

Development and introduction of new antibacterial materials and approaches to treatment and correction of inflammatory processes caused by opportunistic pathogenic microorganisms, and search for the antibacterial materials that may in a number of cases become an alternative to antibiotic treatment, also remains today an issue of primary importance. The purpose of this work has been to study the dominating agents of diseases of the upper airways and determine the antibacterial effect of essential oils upon the clinical isolates.

The research was performed on the basis of the Microbiological Laboratory of the Department of Genetics, Plant Physiology and Microbiology, Uzhhorod National University, and the Bacteriological Laboratory of the Svaliava Central Rayon (District) Hospital.

The analysis of the sputa of 123 patients with inflammatory diseases has shown that in 26 cases bacteria of *Staphylococcus* genus were isolated. Bacteria of *Staphylococcus* genus were the etiological factor of the airways disease in 34%, 12% and 28% cases in 2015, 2016, and 2017, respectively.

Our study showed the level of antimicrobial activity of essential oils against staphylococcus to gradually decrease in the row of *Thymus vulgaris* L., showing

the most distinguished antibacterial effect, *Hyssopus officinalis* L., *Menta piperita* L., and *Coriandrum sativum* L.

Thus, the results of the in vitro study point at high antimicrobial activity of essential oils against the clinical isolates of staphylococcus. Because of that, essential oils can be considered promising materials that may be included into complex antimicrobial therapy of inflammatory processes caused by opportunistic pathogenic microorganisms. The fact that phyto-materials much less often than antibiotics provoke the formation of resistance in bacteria and microscopic fungi may be considered as a motivation in favour of their use.

Krytsova Maryna¹, Salamon Ivan², Bucko Daniel³

ANTIMYCOTIC EFFECT OF ESSENTIAL OILS AGAINST *CANDIDA* ISOLATES

¹Uzhhorod national university, Uzhhorod, Ukraine;

²University of Presov, Presov, Slovakia;

³Calendula, Co., Nova Lubovna, Slovakia

maryna.krivcova@gmail.com

The problem of microorganisms' resistance to antimicrobial materials grows continually worse. In such conditions, special emphasis is being placed on constant monitoring of circulating poly-resistant strains of nosocomial microorganisms and development of new approaches to antibacterial therapy. At the same time, it is shown that the spread of complications caused by the not susceptible to antibiotic *Candida* genus fungi is presently taking on special significance.

Under such conditions, it becomes especially important to perform research aimed at the search for alternative anti-microbial materials. The sources for such materials are the plants that have for a long time been used in popular and traditional medicine. Essential oils, which are promising anti-bacterial remedies used in cosmetology, medicine, food industry, etc.

To determine the antimicrobial activity of essential oils as test cultures were used the typical strain of yeast from the American Type Culture Collection, USA *Candida albicans* ATCC 885-653; 20 clinical strains *Candida* isolated from the sputum of people with pulmonary disease, which taken antibiotic therapy during long period. Antimicrobial activity was determined using disk diffusion method, using 6 mm sterilized filter paper discs. Cultures of *Candida* were previously grown on the elective nutrient media Sabourand Dextrose Agar (SDA) 30 C 48 h.

The essential oils of the following plants were used: *Rossmarinus officinalis* L., *Thymus vulgaris* L., *Menta piperita* L., *Matricaria chamomila* L., *Hyssopus officinalis* L., (produced by «Calendula», Ľubovňa).

The results of the bacteriological examination of the sputum of patients with pulmonary disease showed that that microscopic *Candida* genus fungi were isolated from the sputum of 71% patients, out of 163 examined. The study revealed the high proportion of resistant strains among *Candida* sp.

According to results, it has been found that essential oils of *Thymus vulgaris* L. have significant antimicrobial activity to *Candida* isolates: zones of growth retardation varied from 32.50 ± 0.50 to 75.00 ± 1.50 mm. What was more, all isolates were sensitive to the given essential oil, even those that were resista

Kyrychenko O. V.

AZOTOBACTER CHROOCOCCUM IS THE EFFECTIVE INOCULANT AT THE SPRING WHEAT PLANTS

Institute of Plant Physiology and Genetics of NAS of Ukraine, Kyiv, Ukraine

azoleki@ukr.net

Azotobacter chroococcum is soil bacteria. It is a plant growth-promoting rhizobacteria (PGPR). *Azotobacter* microorganisms have complex positive agriculturally useful abilities for plants and soil such as the nitrogen-fixation, growth activation, bioremediation et al. *Azotobacter* hasn't toxic effect on the human and animals. These are non-pathogenic microorganisms.

The effect of the pre-sowing treatment of spring wheat seeds by *Azotobacter chroococcum* T79 (monoinoculation) as well as by biological compositions created on the basis of *A. chroococcum* T79 strain (complex inoculation) on physiological parameters of development and plant productivity as well as the number and functional activity of rhizospheric diazotrophic microorganisms were studied in greenhouse and field experiments. The strain of *A. chroococcum* T79 was selected from soil of Ukraine in Department of Symbiotic Nitrogen Fixation of Institute of Plant Physiology and Genetics of NAS of Ukraine.

The efficacy of the action of monoinoculation suspension was shown as intensification at plant growth, plant vegetation part formation, chlorophyll and sugars contents in leaves and wheat yield by 10 %. It was shown also that pre-sowing seeds bacterization by complex biological compositions of bacterial (*A. chroococcum* T79 strain and *Agrobacterium radiobacter* 204 strain) and lectin-bacterial nature (*A. chroococcum* T79 strain and wheat germ agglutinin as biological active substance) has an overall positive effect on the components of

«plant–soil–microorganism» system, resulting in wheat crop yield increase on 17–20 % followed by the improvement of microbiological soil characteristics due to the active growth and nitrogen-fixing ability of agriculturally useful microflora. The advantages of binary compositions use as compared to the monocultures were established demonstrating their higher stability in natural agrophytocenosis.

Lazar E.P., Sharga B.M.

APPLE SCAB IN UZHGOROD DISTRICT: 2016-2017

Uzhgorod National University, Uzhgorod, Ukraine

bmsarga@yahoo.co.uk

Apple scab is a most important disease in Transcarpathian orchards. The humid mild climate of our region is suitable for the development of fungi, particularly, *Venturia inaequalis* (Cooke) Wint.

Last two years we are evaluating the apple scab in small and commercial orchards in our region. We take into comparative evaluation 50 small private orchards untreated by fungicides, situating near houses and 2 large commercial orchards, treated by the fungicides Luna Sensation at bloom and by Delan 14 day after the flowering. Symptoms severity grade was applied to evaluate the disease on leaves and also disease agent interaction with different cultivars.

The frosts at the start of year 2016 were strong enough and following summer was hot and dry, the autumn was warm and dry in our region. This resulted in less severe apple scab symptoms, than in year 2017, when growing season was warm and humid.

We found that most severe symptoms were developed onto untreated leaves in small gardens near houses. The most susceptible cultivars were Golden delicious, Champion, Jonagold, Gala, Fuji. The less susceptible in untreated conditions were cultivars Braeburn, Granny Smith, Pinova. Weak symptoms with little spread within the canopy were on resistant Luna, Sirius, Red Topaz, Orion and Rossella trees.

Mentioned above treatment was not sufficient to prevent disease on susceptible and less susceptible cultivars. However, when resistant trees were sprayed, no symptoms developed.

We discovered few rural cultivars with good horizontal resistance to the scab disease agent also.

Lebed A.P.¹, Furtat I.M.¹, Tomina V.V.², Vaclavikova M.³, Melnyk I.V.^{2,3}

**THE ROLE OF SILICA FUNCTIONALIZED MICROSPHERES IN THE
FORMATION OF BIOFILM BY *STAPHYLOCOCCAL* CLINICAL ISOLATES**

¹National University of Kyiv-Mohyla Academy, Kyiv, Ukraine;

²Chuiko Institute of Surface Chemistry of NAS of Ukraine, Kyiv, Ukraine;

³Institute of Geotechnics SAS, Kosice, Slovak Republic

anastasia.lebed3@gmail.com

Biofilm is a complex consortium of microbial cells which is surrounded by the extracellular matrix. This form of existence is believed to be a specific kind of the mechanism of resistance against microbicidal compounds. It provides pathogens with the ability to cause chronic and acute forms of infection. In this regard, the study aimed to investigate the promising method of coping with pathogenic bacteria, which is inhibition of the biofilm formation by cutting-edge antimicrobials - nanoparticles. To investigate the ability to form biofilm in the presence of Cu²⁺-containing silica nanoparticles and those without Cu²⁺, the standardized suspensions of the cells of 18-hour cultures of two isolates, *Staphylococcus sp.* R1 and R2, have been used. It has been achieved with the help of the microtiter assay with a 1% solution of crystal violet. Optical density has been measured at 538 nm. The ability of intact cells to form biofilm has been taken as 100 percent and used as a positive control. Impact of nanocomposites in experimental groups was calculated, comparing to the control one. Isolate R2 has been characterized moderate ability to form biofilms when isolate R1 has performed weak biofilm forming activity. Previously, it has been found that Cu²⁺-containing nanoparticles rather than without them would have more significant antibacterial activity. In this regard, two concentrations of microspheres which have partial bactericidal properties - 0.1 and 0.01% - have been used. After the 120-minute contact of bacterial cells with Cu²⁺-containing nanoparticles, more sufficient inhibiting effect than with those without Cu²⁺ has been found. Thus, the level of inhibition in isolate R1 was 38,1 and 41,43 with the presence of 0.1 and 0.01% solutions of nanoparticles, and in isolate R2 – 42,2 and 22,1. It has been shown that such the essential process would be efficiently blocked in the presence of the silica nanoparticles without Cu²⁺. Apparently, inhibition of biofilm formation, increased amount of nanocomposites and their bigger size would reduce the ability to block sites of adhesion of the bacterial cells.

Levchenko A. G.**HEALTHCARE SYSTEMS ACCORDING TO THE CONCEPTION 'ONE HEALTH
IN UKRAINE'**

Odesa State Agrarian University, Ukraine

AnnLevchenko22.12@gmail.com

Medicine and Veterinary Medicine are both similar and very different areas of knowledge. So, a common problem is to unite efforts of vets and medics in extreme situations, connected with zoonotic epidemics.

The One Health conception came from the recognition of linkages between human and animal health, that's why a holistic approach is needed to understand, to protect, and to promote the health of all species. Whether it is emerging infections disease, antibiotic resistance, globalization, natural disaster, or climate change, human and veterinary medical communities should work together to overcome successfully the serious health threats of the 21st century.

Healthcare systems are complexes for controlling equally individual and public health problems, which are able to intervene on epidemiological processes and reduce the prevalence of diseases through effective prevention and evidence-based treatment strategies.

Each of these measures doesn't work well in the Ukrainian health care system. The rate of tuberculosis infection is more than 30 times higher in Ukraine than in the US. These are long-standing realities that have defined Ukrainian healthcare for decades, because both medics and vets work separately.

One Health tries to improve the well-being of all species by enhancing collaboration between physicians, veterinarians, researchers and by management promotion to achieve these goals through:

1. Joint educational efforts of medical and veterinary schools;
2. Joint communication efforts in journals, at conferences, and via allied health networks;
3. Joint efforts in clinical care through the assessment, treatment and prevention of cross-species disease transmission;
4. Joint cross-species disease surveillance and control efforts in public health;
5. Joint efforts in better understanding of cross-species disease transmission through comparative medicine and environmental research;
6. Joint efforts in the development and evaluation of new diagnostic methods, medicines and vaccines for the prevention and control of diseases across species;
7. Joint efforts to inform and educate political leaders and the public through accurate media publications.

Litvinov S.V., Potrokhov A.A.

**ISOLATION OF *AEROMONAS SALMONICIDA* FROM LEAF EXPLANTS OF
NICOTIANA TABACUM L.**

Institute of Cell Biology and Genetic Engineering of the National Academy of Sciences of Ukraine, Kyiv, Ukraine

slitvinov83@gmail.com

Gram-negative bacteria strain has been isolated from the aseptic in vitro culture of *N. tabacum* leaves and seedlings. Explants, containing bacterial cells, had been selected by the marker of kanamycin resistance on a selective medium. At the absence of kanamycin in the medium regeneration of explants did not differ from controls. Control seeds and leaf explants without symbionts lacked resistance to the antibiotic. Intracellular localization of bacterial symbionts had been confirmed by transmission electron microscopy.

Filterable form of bacteria were recovered from the supernatant of leaf tissue, previously sterilized by 1:3 NaOCl (5 min) and fine chopped in PBS buffer. Filtration was performed through a Whatman syringe filter 25 mm GD/X Sterile, 0.2 µm pore size. When cultured on solid MS medium in aerobic conditions reversion to the vegetative cells about 1-2 µm in size was observed. The bacterium is Gram-negative, catalase- and oxidase-positive. Colonies grow on different nutrient media, both solid and liquid, at a temperature up to 35 0 C with optimum between 22 and 25 0 C. Vegetative bacteria formed S-type colonies that with the aging of culture dissociate and form R-type or G-phase (intermediate) colonies. S-colony consisted mostly of rod cells (1-2 µm in size), R-colony – mostly of coccoid cells (0.5-1 µm in size).

Bacterial cultures from the aseptic in vitro culture of *N. tabacum* leaves were identified as *Aeromonas salmonicida* by BioMerieux VITEK 2 System Version 07.01 (Institute of Microbiological Research, Kiev, Ukraine). Isolated strain was resistant to penicillin, kanamycin, ampicillin, cefazolin, cefotaxime, but sensitive to ciprofloxacin and to ceftazidime. The antibiotic resistance is likely determined by the plasmid factor as resistance has been lost after treatment of bacteria with ethidium bromide. Cefotaxime and ceftazidime induce an ability of isolated bacteria to invade into bacteria-free *N. tabacum* leaf explants.

It can be concluded, that the latent persistence of bacterial symbionts can influence the results of laboratory experiments, as well as normal and pathological physiology of plants under stressful conditions.

Loboda M.I., Biliavska L.O., Iutynska G.O.

LECTINS BIOSYNTHESIS BY SOIL STREPTOMYCETES – ANTAGONISTS OF PHYTOPATOGENES

Zabolotny Institute of Microbiology and Virology, NAS of Ukraine, Kyiv, Ukraine
marichka20loboda@gmail.com

Plants infectious diseases problem, which leads to crop losses and crop production limitations, is not new, but recently the problem has acquired a global character. Therefore, the creation of highly active bioformulations, based on biologically active metabolites of microorganisms for modern phytosanitary technologies is promising.

For the successful application of metabolic compounds in the plants protection, both the isolation of new producers and a more detailed study of their biosynthetic potential are necessary.

In recent years, the interest in lectins and agglutinins of bacteria has grown. These substances are known to have a significant role in the «pathogen-host» interaction.

The goal of the research was to study the ability of soil streptomycetes to produce lectins and to determine their carbohydrate specificity.

As the objects of the researches were strains *Streptomyces avermitilis* IMB Ac-5015, *S. netropsis* IMB Ac-5025 and *S. violaceus* IMB Ac-5027, isolated from the chernozem and chestnut soils.

In result of conducted researches, haemagglutination activity was detected both in supernatant cultured liquid and ethanolic biomass extract of streptomycetes, which were grown on organic and synthetic medium. The hemagglutination titer varied from 1:4 to 1:64 for the supernatant cultured liquids and from 1:16 to 1:256 for ethanolic biomass extracts. That gives the background to suppose the lectins presence in these substances. It was detected the carbohydrate specificity of lectins to L-fucose, D-galactose, D-glucose, D-mannose within the limits of 4.68-9.32 mM in the test samples in the reaction of hemagglutination inhibition .

The revealed ability of streptomycetes-antagonists to produce lectins with a certain carbohydrate specificity gives backgrounds to expect the participation of these metabolites in the molecular and biochemical phytopathogen- plant interaction.

Lutsenko T.M.^{1,2}

**BIOTECHNOLOGY OF RECOMBINANT INTERLEUKIN-7 HUMAN AND ITS
STANDARDIZATION**

¹National Technical University of Ukraine "Kyiv Polytechnic Institute" them. Igor Sikorsky, Kyiv, Ukraine;

² LLC "UA-PRO-PHARMA", Kyiv, Ukraine

tanywalytsenko@gmail.com

Interleukin-7 (IL-7) is one of the central cytokines of the immune system, which plays an important role in the modulation of T- and B-cell development and T-cell homeostasis. IL-7 is a promising drug for the restoration of the immune system of people against the background of immunodeficient states of various origins, including for cancer diseases, bacterial and viral infections.

From the point of view of its therapeutic potential, there is a significant interest in the development of technologies for the production of a biologically active polypeptide IL-7. The optimal solution in developing the technology of obtaining human IL-7 is to create a recombinant producer for the synthesis of the described cytokine.

In order to ensure the quality, efficiency and safety of drugs obtained by biotechnology, the question arises in standardizing methods for controlling their quality. The peculiarity of the standardization of biotechnological products is that each recombinant drug is individual, and therefore requires a personal approach in the development of quality control methods.

With the help of the methods of analysis considered in the work, it is possible to evaluate such important quality indicators as purity, quantitative content of the active substance and biological activity.

The rational parameters of biotechnology for the preparation and purification of recombinant IL-7 (rIL-7) were substantiated, which allows obtaining the latter with high biological activity in vitro. It has been shown that rIL-7 is accumulated in *E. coli* cells of the strain BL21 (DE3) in the form of body-inclusions; the proportion of the target protein is 15-20% of all bacterial proteins, and the yield is 0,8-0,9 mg / ml of culture fluid.

In the course of the work, adaptation of the method of determining the biological activity of rIL-7 with the use of human peripheral blood mononuclear cells was performed. The results of validation of this methodology based on indicators such as specificity, linearity, correctness and precision proved the possibility of using this method for routine analytical quality control of rIL-7.

Mamenko T.P., Dvorak K.P., Kots S.Ya

**FORMATION OF PROTECTIVE REACTIONS IN SYMBIOTIC SYSTEMS
GLYCINE MAX – *BRADYRHIZOBIUM JAPONICUM* UNDER THE ACTIONS OF
PROLONGED DEHYDRATION**

Institute of Plant Physiology and Genetics, National Academy of Sciences of Ukraine, Kyiv
t_mamenko@ukr.net

The changes in the activity of key antioxidant enzymes, superoxide dismutase, glutathione reductase, ascorbate and guaiacol peroxidase, have been investigated in order to determine the formation of protective reactions in symbiotic systems *Glycine max* - *Bradyrhizobium japonicum* on prolonged dehydration.

The objects of the study were symbiotic systems formed with the participation of soybean (*Glycine max* (L.) Merr.) and nodule bacterial strains (*B. japonicum*) – 646 (active, virulent), 604k (inactive, highly virulent) and Tn5 mutants B1-20 (active, virulent), 107 (low active, virulent).

It is found, that symbiotic systems, formed with the participation of soybean and the active strain *B. japonicum* 646, as well as the active Tn5 mutant B1-20, are differed by insignificant changes in the activity of antioxidant enzymes – superoxide dismutase, glutathione reductase, ascorbate and guaiacol peroxidase in root nodules and root plants during dehydration.

It has been recorded that inoculated of soybean with the low-activity Tn5 mutant *B. japonicum* 107 results in an increase in the activity of the investigated enzymes, especially superoxide dismutase in the root nodules and root plants in the conditions of moisture deficiency, as well as raised activity of glutathione reductase after recovery of irrigation plants.

It is shown that the symbiotic system, formed with the participation of soybeans and the inactive *B. japonicum* 604k, strain leads to differently directed changes in the activity of enzymes – suppression of the activity of superoxide dismutase and glutathione reductase and increased activity of ascorbate and guaiacol peroxidase in nodules and root plants under dehydration, as well as weak activity restoration of all enzymes in after a stressful period.

Conclusion that the formation of protective reactions of soybean plants in symbiosis with *B. japonicum* under the actions of prolonged dehydration is accompanied by adaptive changes in antioxidant enzymes activity and depends from the ability of the symbiotic system to realize its adaptive potential under stress conditions.

Masur T.V., Pronko I.V.

**ANALYSIS OF A PROBABLE DANGERS, CAUSED BY A VIRUSES FAMILY
FILOVIRIDAE**

National University of Life and Environmental Science Kiev, Ukraine

doktorvet67@ukr.net

Among the activators of dangerous anthroponoses diseases, one of the most interesting is family Filoviridae, that includes 7 species of *Ebolavirus*: *Zaire ebolavirus*, *Bundibugyo ebolavirus*, *Sudan ebolavirus*, *Marburg marburgvirus*, *Lloviu cuevavirus*, *Reston ebolavirus*, *Tai forest ebolavirus*. *Zaire*, *Bundibugyo* and *Sudan* are associated with a causes of powerful epidemics. *Reston ebolavirus* is an extraordinary representative of a family. It was found in pigs, that had no clinical signs of the disease, but was very dangerous for primates, and *Lloviu cuevavirus*, that was found in flyings mice in a Spain caves, and is apathogenic for human. That's interesting, that in 1945 pr. Chumakov highlighted the virus (exciter of Crimean-Congo fever), in a result of an outbreak of disease with with high mortality in Crimea. In a course of long-term research, the identity of the genome of the *Ebolavirus* and hemorrhage fever of Crimea-Congo was finally proved. Therefore, the time of infection occurrence, caused by the family of phyloviruses, geographical delimitation and certain genetic differences, show us the need for analysis to detect the natural reservoir of phylovirus, that sometimes is in contact with living organisms, that are sensitive to it, and it can lead to the development of a serious illness with a classical symptom. Until a certain time among the researchers there was no single opinion about the potential role of predatory mammals in a circulation of the *Ebola* virus in the areas of ill-being. However, in the outbreak site, 30% of domestic dogs detected specific antibodies to the virus. Since such animals are characterized by asymptomatic carriers, they potentially serve as a reservoir of *Ebola* virus. The largest value in the role of the reservoir and vector of the *Ebola* virus is the representatives of the family *Viverridae* (*Hemigalinae* and *Genetta*), whose premises are natural hollow structures (hollows, caves, grottoes). Often these biotopes inhabit several generations in succession of different types of bats. Under conditions of joint use of biotopes, optimal conditions for the transmission of phyloviruses through products are formed.

Melnychuk T.M., Egovtseva A.Yu.

**THE INFLUENCE OF FARMING SYSTEMS ON THE MICROBIOCENOSIS OF
SOUTHERN CHERNOZEM**

FSBIS "Scientific Research Institute of Agriculture of the Crimea", Simferopol, Crimea
melnichuk7@mail.ru

Microbial community of soil of agroecosystems depends on anthropogenic load. Implementation of no-till is one of important factors of water- and energy preservation and improvement of soil tolerance to erosion which is important for the southern steppe. In regard to that, the aim of our research was to conduct analysis of microbial community structure in soil southern chernozem under the influence of a microbial preparations complex and different farming systems (traditional, that is based on tilling, and no-till). The numbers of microorganisms of the main ecological-trophic groups of southern chernozem under the conditions of stationary experiment have been learned. The highest number of microorganisms in all studied groups in the upper layer (0-10 cm) of southern chernozem was observed during autumn. It was shown that under the farming systems no-till, the number of oligotrophs and cellulolytic agents of southern chernozem increased, while the number of nitrogen-fixing microorganisms, micromycetes and pedotrophs decreased. A positive effect of seed inoculation by a complex of microbial preparations on the number of microorganisms of the main ecological-trophic groups and tendency to reduce the intensity of mineralization processes was shown. Number of cellulolytic agents of southern chernozem increased in variants with the use of a complex of microbial preparations for 30% under conditions of no-till and more than 2.5 times compare with traditional farming system.

Thus, no-till farming system influenced microbial communities of southern chernozem. A complex of microbial preparations increased the number of soils microorganisms of the main ecological-trophic groups.

Matsas O.U., Mychalchuk G.A., Mulkina O.I., Slobodyanok O.M.

**DETECTION OF *IXODIDAE* ACARI BORRELIOSIS IN KIEV BY MOLECULAR-
BIOLOGICAL METHOD**

Aleksandrovskaia Clinical Hospital in Kyiv

matsas@ukr.net

Among natural focal infections registered in the city of Kyiv, such infections as ixodid tick-borne borreliosis are of great medical and social importance. The presence of large natural and park zones in Kyiv contributes to the spread of

ixodid ticks that carry borrelia, posing a great danger to human health, as the disease often leads to long-term disability.

The purpose of the study was to determine the presence of *Borrelia* in mites. removed from people after a bite in different natural and park areas of Kyiv.

45 ticks of the genus *Ixodes* were delivered to the laboratory of the Aleksandrovskaya Clinical Hospital in Kyiv

A real-time polymerase chain reaction, using a set of reagents to detect 16S rRNA *Borrelia burgdorferi sensu lato* (*B. burgdorferi sensu strict*, *B.afzelii*, *B.garinii*) was used to identify *Borrelia* in biological material.

As a result of the analysis, there was detected a high dissemination of ticks by *Borrelia*. Out of 45 treated samples isolated from ticks, 21 samples gave a positive result to the presence of *Borrelia burgdorferi sensu lato* 16S rRNA, which is 46.6%.

The use of molecular biological methods makes it possible to study natural focal infections in various regions and large megalopolises, and also allows caring out preventive measures against difficult-diagnosed transmissible natural focal diseases.

Detection of an infectious agent in mites is important for the correct diagnosis, timely treatment, and it also reduces patient's psychological stress.

Matviienko N.M.¹, Shepelevych V.V.²

ASSESSMENT OF BACTERIAL MICROFLORA IN FISH OF FRESH WATER AQUACULTURE OF UKRIANE

¹NAAS Institute of Fisheries, Kyiv, Ukraine;

²Taras Shevchenko National University of Kyiv, ESC "Institute of Biology and Medicine", Kyiv, Ukraine

mnarine73@ukr.net

In 2017 in the examined fisheries of Ukraine both autochthonous (or water), and allochthonous microflora in fish was studied. We performed 71 isolates of commensal and pathogenic bacteria, which were included into author collection of the Department of Ichthyopathology of the Institute of Fisheries of NAAS.

The isolates were obtained from 10 fish species of various age categories. In particular, we performed 17 isolates from branchiurans (namely, 12 from carp and 5 from koi), 9 from salmonid fishes, 17 from acipenserids, 17 from trout, and 12 isolates from other fish species (catfish, tilapia, roach, pike perch, and pike).

We performed the isolates primary identification, and study of their morphological and biochemical properties.

Water temperature is one of the leading factors in microorganisms' development, causing the content of microorganisms. The highest values of microflora quantitative content were registered in June, July, as pathogenic microflora (vibrioflora) and opportunistic microflora (*Aeromonas*, *Pseudomonas* and *Enterobacteriaceae*).

In the fish studied during cold seasons dominated the psychrophilic bacteria of the species *Pseudomonas*, *Aeromonas*, and *Flavobacterium*. During warm seasons the microflora of skin and internal organs in fish was represented by mesophilic microorganisms – various bacteria of genus *Micrococcus*, corynebacteria. We also isolated the representatives of genus *Proteus* (*Proteus morganii*) and a representative of *Enterobacteriaceae* family - *Escherichia coli*, as representative microorganisms of the sanitary condition of the polluted waters. The most common isolated microflora were opportunistic bacteria of genus *Pseudomonas*, *Aeromonas*, and *Flavobacterium*; in small quantity we identified the representatives of *Enterobacteriaceae* family, and the small numbers of bacteria of genus *Staphylococcus*, *Streptococcus*, and *Proteus*.

In particular, the bacteria of genus *Aeromonas* and *Flavobacterium* in the majority of cases were isolated from fish juveniles and this year rainbow trout (*Oncorhynchus mykiss*), of genus *Pseudomonas*, *Aeromonas* - from cyprinoids, namely from carp (*Cyprinus carpio*), and the bacteria of *Yersinia* genus from trouts.

In gastrointestinal tract of fresh water fish the genus *Enterobacter*, and *Aeromonas* predominated. We also registered *Escherichia*, *Klebsiella*, *Proteus*, *Serratia*, *Staphylococcus*, and *Pseudomonas*.

Nastenko V.B., Osypchuk N.O.

FEATURES OF DETECTION OF MRSA STRAINS AMONG CLINICAL ISOLATES

Bogomolets National Medical University, Kyiv, Ukraine

encelad1991@gmail.com

In a 1945 interview with The New York Times, Alexander Fleming predicted appearance of penicillin resistant bacteria. This forecast confirmed within 10 years of the widescale introduction of penicillin. The creation of methicillin was the panacea. It happened in 1960. However, thereafter, strains of *Staphylococcus spp.* were allocated that were resistant to this drug. According to the CDC, there were more than 80,000 invasive MRSA infections and 11,285 related deaths in

2011. The purpose of the study was to identify MRSA strains among clinical isolates of *Staphylococcus spp.*

The study involved 45 clinical strains of *Staphylococcus spp.* The sensitivity to antibiotics was determined relative to oxacillin (an analogue of methicillin) and vancomycin (it has been extensively used for treating MRSA infection). We were using agar dilution method ("wells method" for vancomycin and disk-diffusion method for oxacillin). The disks with oxacillin contained 1 µg of active compound. The vancomycin was introduced into the wells of agar in the amount of 20 µl (in concentration of 1 mg in 1 ml). Clinical isolates with growth zones of delay around the disk with oxacillin retardation up to 10 mm were considered to be methicillin-resistant (according to the order 167). The vancomycin-resistant strains were those who formed growth zone less than 15 mm.

As a result of experiment, it was found the sensitivity clinical strains of *Staphylococcus spp.* to oxacillin and vancomycin. Sixteen strains were defined like MRSA; it is 35,56% from all studied strains. The oxacillin formed growth zones in range of 10 to 20 mm relative to three strains (6,67%). Most of the test-microorganisms were too sensitive to the drug (57,78%). In most cases, vancomycin has caused formation of growth zones with a diameter of more than 15 mm, but five strains were determined like resistant to vancomycin.

The results of an experimental study show that 35,56% of strains of *Staphylococcus spp.* were determined like MRSA. It proves, that MRSA remains an important public health problem and more remains to be done to further decrease risks of developing these infections.

Nazarchuk O. A., Nahaichuk V.I., Osadchuk N. I.

**ANALYTIC PROGNOSIS OF THE SUSCEPTIBILITY TO β-LACTAM
ANTIBIOTICS IN *A. BAUMANNII* AND *P. AERUGINOSA*, CAUSING
INFECTIOUS COMPLICATIONS IN PATIENTS WITH BURNS**

National Pirogov Memorial Medical University, Vinnytsya Ministry of Health of Ukraine,
Vinnytsya, Ukraine

nazarchukoa@gmail.com

Acinetobacter baumannii, *Pseudomonas aeruginosa* are widely known to be of predominant significance in etiology of infectious complications among patients with burn trauma.

Aim – to carry out analytic prognosis of susceptibility to β-lactam antibiotics in clinical strains of *A. baumannii* and *P. aeruginosa*, isolated from patients with burns.

Materials and methods. In microbiological observation (2011-2017) clinical strains of *A. baumannii* (n=224), *P. aeruginosa* (n= 127) were isolated from 510 patients with burns. Patients with 2nd-b-3rd degrees of burns underwent standard administrations accordingly to standard protocols in Vinnitsa Regional Clinical Hospital named after N. I. Pirogov. Clinical strains of *A. baumannii*, *P. aeruginosa* were isolated and identified by means of standard microbiological methods. The sensitivity of *A. baumannii*, *P. aeruginosa* to β -lactams (ampicillin/sulbactam, amoxicillin/clavulanate, ceftazidime, cefoperazon, cefoperazon/sulbactam, meropenem) was studied by standard disc-diffusion and quantitative serial dilution tests. The analytical prognosis of the effectiveness of β -lactams against *A. baumannii*, *P. aeruginosa* isolates was obtained, using mathematical prognostication for conducting prognostic mathematical models ("STATISTICA 7"; "Matlab 7.11").

Results and discussion. The analysis demonstrated decreasing tendency of the sensitivity in *A. baumannii*, *P. aeruginosa* to amoxicillin/clavulanate (25-42 %). In *A. baumannii* we proved the recovery of their prior low (16 % and 34 %) susceptibility to ampicillin/sulbactam and cefoperazon/sulbactam to the rates higher than 70 % (in 2017). The susceptibility of both bacteria to ceftazidime was described alike parabolic function with prior recovery and further decrease (lower 25,9 %). We found low effectiveness of meropenem against *A. baumannii* (25 %) alike sinusoid function and alike exponential decrease in *P. aeruginosa* (78 % to 30 %).

Conclusion. The analytical prognostic formulas express decreasing susceptibility of *A. baumannii* and *P. aeruginosa* to the majority of β -lactams. The recovery of ampicillin/sulbactam and cefoperazon/sulbactam effectiveness was found only in *A. baumannii* isolates.

Oriabinska L.¹, Gorchakov V.¹, Prasanna D. Belur²

THE POTENTIAL BIOTHERAPEUTIC EFFECT OF *LACTOBACILLUS*
PLANTARUM 2621

¹National Technical University of Ukraine "Igor Sikorsky Kyiv Polytechnic Institute", Ukraine;

²National Institute of Technology Karnataka, Surathkal, Mangalore
olanab9@gmail.com

Probiotic strains with antioxidant properties offer a new perspective for the fight against diseases caused by oxidative damage in the human body. Especially, strain *L. plantarum* 2621 from the collection of microorganisms of the Institute of Microbial Technology IMTECH, India is more promising. This strain produce tannase, which acts upon dietary tannins to produce gallic acid which is

an excellent antioxidant. There are reports on the ability of gallic acid to protect cells from oxidative damage and inhibit the malignant process of the tumor in animal models. Thus, the search for tannase-positive strains and the development of probiotic formulation with a desirable therapeutic effect is an interesting proposition. The aim of the study was to determine the biotherapeutic properties of the *L. plantarum* 2621. The biotherapeutic effect of strain was assessed using interactive spectral-dynamic analysis (SDA) on the base medical expert complex (CME). The CME includes the programmes: neurology, oncology, gastrointestinal tract, genitourinary system, cardiovascular system: orthopedics, endocrinology, otorhinolaryngology, dentistry, ophthalmology, pulmonology, dermatology. According to the CME principle, tissues and organs have a certain vibrational frequency spectrum, including cellular torsion fields. A comparison of the vibrational spectra of probiotics and macroorganism makes it possible to predict their effect on the disease.

The analysis showed that biotherapeutic effect of culture was the highest (up to 80% of similarities in the spectra) for 32 diseases of the gastrointestinal tract and 49 oncological. The similarity of spectra with 28 female and 21 male diseases of the urinary tract, 10 stomatological, 25 endocrinological and 10 dermatological diseases was established.

High indicators of the biotherapeutic activity of the *L. plantarum* 2621 showed the prospects of further studying the probiotic and technological properties of the strain for developing a probiotic drug based on it.

Osypchuk N.O., Nastenko V. B., Ponyatovskiy V.A.

LOCATIONS FEATURES OF GENUS *CANDIDA* REPRESENTATIVES IN ORAL BIOTOPES IN CANCER PATIENTS

National medical University, Kyiv, Ukraine

OsypchukNO@i.ua

Introduction. Fungi of the *Candida* are the resident microflora of the oral mucosa with high carrier level. Candidiasis is a common and serious complication of cancer and its therapy.

Materials and methods. We have examined the composition of different oral mucosa biotopes concerning its *Candida* fungi aspect in 50 patients with cancer. The microbiological material was collected from mucous membrane of the cheeks (retromolar are), dorsal surface of the tongue, mouth angle (mucosa and skin border), hard palate mucous membrane. In the paper bacterioscopic, mycological, statistical research methods were used. Identification of *Candida*

yeasts was performed by using a test – ID of 32 test strips and BioMerieux Company HiCrome Candida Agar/Modified.

Results. By results of the conducting studies fungi of the genus *Candida* on the mucous membrane of the oral cavity patients with cancer were found in 68% (34 patients) of individuals. Moreover, 29 patients – 58% of patients had candidiasis of the oral cavity – quantitative composition of yeast fungi of the genus *Candida* was in the range of 10^4 – 10^6 CFU (colony forming units). Other 5 persons – 10% of the patients belonged to candida-carriage – number of *Candida* was less than 10^3 CFU.

In representatives of the genus *Candida* in the oral cavity within biotopes *C. albicans*, *C. krusei*, *C. tropicalis*, *C. glabrata* were revealed. Thus 75,3 % is *C. albicans*, 12% – *C. krusei*, 1,1% - *C. tropicalis*, 9% - *C. glabrata*.

Conclusion. *Candida albicans* is the most common representative among identified *Candida*'s in all investigated oral biotopes (75,3%).

Pareniuk O.¹, Simutin I.², Samofalova D.³, Shavanova K.¹

IN-SILICO ESTIMATING METRICS FOR ALPHA AND BETA DIVERSITY OF MICROBIOME

¹National University of Life and Environmental Sciences of Ukraine, Kyiv;

²Taras Shevchenko National University of Kyiv, Ukraine;

³GA Institute of Food Biotechnology and Genomics of NAS of Ukraine, Kyiv
olena.pareniuk@gmail.com

The study of the radioactive substances dynamics, changes in ecosystems, caused by it is the main objective of modern radioecology, which will allow moderating the migration of radionuclides in the environment. Detailed data, obtained from NGS analyzes, makes it possible to study the changes in ecosystems caused by the variation of ecological factors at the ecosystem level and more accurately predict the effects of anthropogenic impacts, and therefore can to be an ideal instrument for assessing the impact of radioactive contamination.

Bioinformatic analyses can influence significantly on the quality of the received. Thus, an incorrect analysis may lead to an incorrect interpretation and, therefore, a low reliability of the data received. This work proposes a method for assessing alpha and beta diversity of microbiome, which was tested on samples taken from the sarcophagus over the destroyed 4th power unit of the ChNPP. QIIME, open source, Phyton-based bioinformatics software, was used to conduct all calculations.

Alpha diversity stands for the biodiversity (number of taxa) within a single community and calculate the amount of detected taxonomy in a particular sample. We used *chao1*, *shannon*, and *dominance* metrics to evaluate alpha-diversity using *alpha_diversity.py* with *.biom and *.tre input files.

Beta diversity evaluates diversity between multiple communities. *Bray_curtis*, *chisq*, *euclidean*, and *parameters* were used to describe it. The QIIME module *beta_diversity.py* was used, which requires the *.biom table entry and metric parameters to be entered. To calculate the beta variety, a merged *.biom table (merged) containing *.biom tables of all samples were applied. Visualization of beta diversity was done using PCoA charts using the QIIME module *principal_coordinates.py* and *make_2d_plots.py*.

Pastoschuk A. ¹, Butsenko L. ², Skivka L. ¹, Patyka V. ²

THE EFFECT OF *PSEUDOMONAS SYRINGAE* PV. *CORONAFACIENS* ON
THE GERMINATION OF WHEAT SEEDS OF DIFFERENT VARIETIES

¹Taras Shevchenko national university of Kyiv, Ukraine;

²D.K. Zabolotny Institute of Microbiology and Virology, NASU, Kyiv, Ukraine
kotsyuk93@ukr.net

Today, diseases of cereal crops caused by *Pseudomonas syringae* are distributed in all regions of Ukraine. *Pseudomonas syringae* pv. *coronafaciens* is causative agent of halo blight and brown bacteriosis of rye. The most intensive development of the disease is observed during the period of grain ripening. The main source of the infection is contaminated seeds. The pathogen is stored in an infected seed for up to three years. In the nature, *Pseudomonas syringae* pv. *coronafaciens* attacks mainly rye and oats but can infest wheat through artificial infection. Meanwhile, widely spread causative agent of basal bacteriosis of wheat in Ukraine is *P. syringae* pv. *atrofaciens*.

This study was aimed to investigate the effects of alive and warmed *P. syringae* pv. *coronafaciens* as well as lipopolysaccharide (LPS) of these bacteria for the germination of wheat seeds of *Discus* and *Pecheryanka* varieties and comparison of their effects with the action of conventional causative agent of basal bacteriosis of wheat.

Wheat variety *Pecheryanka* is moderately sensitive to the infection caused by *P. syringae* pv. *atrofaciens* in comparison with high receptive variety *Khutoryanka*. Exposure to alive cells of *P. syringae* pv. *atrofaciens* 9400 was accompanied by the decrease of wheat seed germinating capacity of *Pecheryanka* variety by 15%, and *Khutoryanka* variety – by 20%. The length of

the roots of wheat seedlings of Pecherianka variety after treatment with suspension of living cells *P. syringae* pv. *atrofaciens* was decreased by 10% in comparison with control, whereas in Khutoryanka variety – by 40%. The pathogenic effect of *P. syringae* pv. *coronafaciens* 9030 was comparable to that of *P. syringae* pv. *atrofaciens* 9400. It has been found that the treatment of wheat seeds of Pecheryanka variety with *P. syringae* pv. *coronafaciens* 9030 results in a decrease its germinating capacity by 15%. Meanwhile, germinability of another wheat variety – Discus - was decreased by 10%. The treatment of wheat seeds with lipopolysaccharide (LPS) from *P. syringae* pv. *coronafaciens* 9030 also led to a decrease of wheat seed germinability.

Thus, *P. syringae* pv. *coronafaciens* 9030 and its LPS decrease germinating capacity.

Patyka V.P.

BIOLOGICAL NITROGEN AND BIOPROTECTION IN THE SYSTEM OF NATURAL AGRICULTURE

DK Zabolotny Institute of Microbiology and Virology National Academy of Science of Ukraine, Kyiv, Ukraine

patykavolodymyr@gmail.com

The problem of biological fixation of nitrogen, along with the problem of photosynthesis, is the basis for the origin and maintenance of life. One of the striking examples of the highly effective use of this unique phenomenon includes USA. Of the 22 million tons of nitrogen used in the agrarian sector, 12 million tons are the "biological" nitrogen. Unfortunately, in our country this problem is not solved satisfactorily, and as a result the fertility of the soil decreases, and so does the productivity of plants, while the ecological situation worsens. In 2017, the bean component in the agriculture system of our state was about 5.8%, whereas in the highly developed countries it is not less than 20%. At the same time leguminous plants are a non-alternative component of the agrosphere crucial for maintaining the soil fertility and solving the protein problem.

In Ukraine, biopreparations such as risobofit, risoagrin, rizoenterin, flavobacterin, agrofil, diazobacterin were created for leguminous, cereal, and vegetable crops by scientists of the Ukrainian Academy of Agrarian Sciences and the National Academy of Sciences.

Bioprotection is yet another important issue.

Despite significant achievements due to the widespread use of pesticides, in particular for control of plant pests and carriers of numerous human and animal

infections, scientists and the public are alarmed by the problem of residues of pesticides in the environment, food, feed, raw materials and water.

These residues, even in small quantities, might be a threat to humans, domestic and wild animals. Metabolism and natural disintegration of pesticides, their movement in the external environment have not been extensively studied.

Biological agents, including microbiological ones, are the potential alternatives to synthetic pesticides, which are distinguished from the chemicals by their much greater selectivity of action, and, therefore, environmental safety.

Biological protection of plants fully meets the requirements of the recently emerging global issue of biologization of agriculture and plant growing. In many countries, the course is aimed at the production of food products without chemical agents. Supermarkets appeared selling ecological / organic products that are in great demand.

Peretiazhko I.A.¹, Voychuk S.I.²

ROLE OF THE PPN1 AND PPX1 POLYPHOSPHATASES IN THE PROCESS OF INTERCELLULAR INTERACTIONS

¹Taras Shevchenko National University of Kyiv, ESC "Institute of Biology and Medicine";

²Zabolotny Institute of Microbiology and Virology National Academy of Sciences of Ukraine
svoychuk@hotmail.com

Biological adhesion is an important ecological property of microorganisms that depends on the structural features of microbial cells surfaces. The structure of any biological surface is a result of work of various energy-consuming intracellular processes among which a unique role is played by phosphorus compounds. In yeast, there are two main enzymes that involved in the metabolism of such compounds: the vacuolar endopolyphosphatase PPN1 and the exopolyphosphatase PPX1. The physiological and biochemical role of these enzymes is still under study and it is known that violation of their biosynthesis leads to changes in the content and length of various polyphosphate chains in various cellular compartments. The aim of this work was to determine the participation of these enzymes in the yeast-mammalian interactions.

Yeast *S. cerevisiae* defective in the genes of polyphosphatase PPX1 and PPN1 and mammalian cells L-929 (mouse fibroblasts), TPC (testicular piglets cells) and MDBK (bovine kidney cells) were used in the study. Mammalian cells were grown to form monolayers and after washings with PBS the yeast cells were added 1:100. Samples treated at 37°C for 60 min. The quantity of nonadherent yeast cell was determined by the colony forming unit, whilst adherent ones were fixed and

stained with acridine orange to detect cytomorphological changes of mammalian cells.

Shown that yeast adhesion to mammalian cells depended on the deficiency of PPN1 and/or PPX1. The presence of potential adhesion blockers influenced yeast adhesion to various degrees and let to reveal additional differences in adhesion between parental and mutant yeast strains. The optical properties of mammalian cells changed as a result of their interaction with yeasts and this was related to the apoptotic/necrotic processes initiated in them in the presence of yeasts, and this phenomenon was amplified in case of the yeasts defective in polyphosphatases. Therefore the lack of polyphosphatases PPN1 and PPX1 influences yeast adhesion to mammalian cells through the changes of the surface properties and synthesis of some extracellular metabolites, which possess cytotoxic activity against the mammalian cells.

Pohilko Yu. M.

**INFLUENCE OF BACTERIA OF THE GENUS *LACTOBACILLUS*, ON THE
PRODUCTIVITY OF YOUNG RABBITS**

Institute of Agricultural Microbiology and Agroindustrial Manufacture NAAS

pohilko.yura@gmail.com

The urgent task of industrial livestock farming is to develop ways of treating and preventing opportunistic intestinal infections, which may be an alternative to the use of antibiotics. The promising direction is the use of probiotics. Most often, these drugs include lactic acid bacteria, which have a pronounced antagonistic activity to pathogenic and opportunistic microorganisms. Probiotics, which are used to eliminate the problems of the gastrointestinal tract of rabbits, have in their composition bacteria isolated from various ecological niches. In connection with this, the creation of a probiotic preparation based on biologically active representatives of the obligate microbiota of the gastrointestinal tract of these animals remains topical. It should be noted that the number of lactic acid bacteria in the gastrointestinal tract of rabbits exceeds the number of bifidobacteria, regardless of diet and type of feeding. That is why a promising group of microorganisms for creating probiotic preparations for rabbits are bacteria *Lactobacillus*, which have the status of "GRAS" and "QPS", that is, their use is absolutely safe.

A study of the effect of bacteria of the genus *Lactobacillus* on rabbits during weaning was conducted on the rabbit farm "Rabbitsland" which is located in Chernigov, Ukraine. Experiments were conducted on rabbits of Poltava silver at

the age of 30-35 days ($n = 18$). The bacterial suspension was added to the drinking water. A strain of *L. helveticus* 13/2 bacteria isolated from the gastrointestinal tract of rabbits was used.

As a result of the studies, there was no significant difference in the weight of the animals of the experimental and the control groups during the entire period of cultivation of rabbits. However, it should be noted that the introduction of a bacterial suspension in the diet of feeding allowed to reduce the feed costs by 10% per 1 kg of weight in the research group's rabbit, indirectly may indicate better assimilation of nutrients from the feed in the experimental group.

The data obtained by us allow us to recommend the strain of *L. helveticus* 13/2 to create a probiotic preparation based on it.

Pronina O.V.^{1,2}, Rushkovsky S.R.², Morgun B.V.¹

**INFLUENCE OF MITOCHONDRIAL DNA LOSS ON DYE ACCUMULATION IN
AGING COLONIES OF YEAST *SACCHAROMYCES CEREVISIAE***

¹Institute of Cell Biology and Genetic Engineering;

²Taras Shevchenko National University of Kyiv, Ukraine

olpronina@gmail.com

Yeast *Saccharomyces cerevisiae* is widely used as a model to study influence of mitochondria state on cellular aging, but information about impact of mtDNA loss on this process in complex yeast population is controversial. Addition of dyes to nutrient media permits to differentiate dead cells in population and thus distinguish areas in which dying cells are concentrated. The aim of our study was to investigate the influence of mitochondrial DNA loss on accumulation and distribution of dyes in aging yeast colonies.

The work was conducted on *S. cerevisiae* diploid strain SK1 and its ρ^0 clones. Yeast cells were inoculated in rich YPD medium with addition of dyes (0,01% bromocresol purple or 0,0004% floxin B) and cultivated at 28°C for 14 days. Colony forming ability was determined by microcolony growth analyses while dye accumulation in cells was verified by luminescent microscopy.

By the third day of cultivation on the dyes containing medium, we registered the appearance of light coloration in the central part of the parental strain colony. Upon further cultivation, it spread throughout the entire surface of the colony with the exception of narrow uncolored edges. The staining of the ρ^0 clone at the initial stages of growth resembled the accumulation of dye in the parental strain, featuring some asymmetric small areas of intensive coloring appeared in the colony center. Later on, some clearly defined zones of intense coloration were

formed in the center of the colonies, which gradually increased as the colony aged. The evaluation of cell state in the middle of the petite colonies confirmed significant loss of their viability ($5,3 \pm 0,9\%$ viable cells after 14 days). They accumulated dyes and were incapable of forming microcolonies on fresh medium. In contrast, more cells from the center of the ρ^+ colony preserved their viability ($29,7 \pm 2,4\%$ viable cells after 14 days), and their coloration was largely determined by the accumulation of dye in ascospores.

Our observation suggest that loss of mtDNA results in dyes accumulation in the central area of petite colonies grown on YPD medium because of decrease of ρ^0 yeast cells viability due to accelerated aging.

**Pysmenna Y., Panyuta O., Belava V., Batsmanova L.,
Kondratiuk T., Taran N.**

**THE USING OF *BACILLUS SUBTILIS* BACTERIAL ISOLATES FOR
ACTIVATION OF WHEAT SEEDLINGS PROTECTIVE REACTIONS UNDER
BIOTIC STRESS**

Taras Shevchenko National University of Kyiv, Ukraine

pismennaya64@gmail.com

The treatment of seeds or plants by endophytic bacteria isolates activates plant protective reactions under stress. The change of lectin activity (LA) and intensity of lipid peroxidation (LPO) refer to the main plant reactions to stressors. That is why the aim of our work was the study of *Bacillus subtilis* 537/B1 bacterial isolates effect on change of LA and intensity of LPO in wheat seedlings under infection by eyespot causal agent *Pseudocercospora herpotrichoides* (Fron) Deighton.

Experimental variants – 7-day-old wheat seedlings: control; infected with a fungal conidia suspension (CSF); infected with a suspension of bacterial isolates (CSB); infected with CSF+CSB; from seeds inoculated with a CSB; from seeds inoculated with CSB and infected with CSF.

Two winter wheat varieties were used – Myronivska 808 and Renan. LA was determined by erythro-agglutination assay. Protein content was determined by the Bradford method. The level of generated malonic dialdehyde (MDA) as product of LPO was estimated according to Dhindsa and Matowe.

Our results at 48 hours after treatment showed the increase of LA in Myronivska 808 experimental variants compare to control. In the Renan seedlings there was no significant difference compare to control, except for the variant infected with CSF + CSB.

Thus, the treatment with *Bacillus subtilis* caused the increase of Myronivska 808 seedlings LA. For this variety the seedling treatment was more effective than seed treatment. Similar reaction was not observed for Renan seedlings at 48 hours after treatment.

Treatment with CSB in both variety seedlings infected by *P. herpotrichoides* caused the reduction of LPO intensity compare to control at 48 hours after treatment. The seedlings of resistant variety Renan showed a lower level of MDA compare with seedlings of susceptible variety Myronivska 808.

Consequently, our research suggests that *Bacillus subtilis* bacterial isolates could be used to activation of plant protective reactions under the action of biotic stressors.

Pysmenna Yu.B., Chuienko A.I.

BIODEGRADATION OF PLASTERBOARD BY MICROMYCETES

D.K. Zabolotny Institute of Microbiology and Virology of the National Academy of Sciences of Ukraine

ulitca@ukr.net

Fungi thanks to a number of morphological, physiological and genetic especially occupy a dominant position among the organisms that cause damage to a number of technical substrates. So it is important to study the characteristics of microscopic fungi on building materials, including drywall, and development of effective measures against this phenomenon. The aim was to investigate the growth of micromycetes on plasterboard and determine their enzymatic activities.

Methods. To study the localization of microscopic fungi in the inner layers of plasterboard used confocal laser scanning microscope. Enzymatic activity (amylolytic, cellulolytic, lipolytic activities, oxidase and catalase) were determined according to size of area of enzymatic activity on solid agar media and enzymatic index. **Conclusion.** It is shown that the most intensive growth of the mycelium of micromycetes was observed on the surface of cardboard and gypsum of plasterboard. It was found that micromycetes isolated from the plasterboard have mostly moderate amylolytic, cellulolytic and lipolytic activity. Catalase activity was detected in *Aspergillus niger* and *Trichoderma viride*, peroxidase - in *Aspergillus flavipes*, *A. niger*, *Chaetomium globosum*, *Neosartorya fischeri*, *Stachybotrys chartarum* and *T. viride*. Micromycetes damage plasterboard not only by colonization of cardboard and growth inside the gypsum core. Found that micromycetes isolated from the plasterboard have moderate and high enzymatic activity, which may explain their intensive growth on this material. Thus,

Aspergillus versicolor F-41226 characterized by high amylolytic and cellulolytic activities. *Cladosporium cladosporioides* F-41230, *C. sphaerospermum* F-41232 had a high lipolytic activity.

**Rudneva I.I.¹, Rudyk M.P.², Shepelevych V.V.², Skivka L.M.², Roslova N.N.²,
Chesnokova I.I.¹, Kovyrshina T.B.¹, Shaïda V.G.¹**

**COMPARATIVE STUDY OF HEALTH BIOMARKERS OF TWO BLACK SEA
TELEOST FISH SPECIES *SCORPAENA PORCUS* AND *SYMPHODUS TINCA***

¹Kovalevski Institute of Marine Biological Research, Crimea

²Taras Shevchenko National University of Kyiv, ESC "Institute of Biology and Medicine",
Kyiv, Ukraine

svg-41@mail.ru

Fish are good indicators for the evaluation of environmental health. Their histological and biochemical parameters are used as biomarkers in many ecological studies and monitoring programs as diagnostic instruments for the analysis of marine waters status. At the other side, different fish species demonstrate different resistance and sensitivity to environmental stress, caused anthropogenic pollution. The aim of this study was to compare the response of two Black Sea teleost fish species benthic *Scorpaena porcus* and benthic/pelagic *Symphodus tinca*, collected in Sevastopol bays, to the contamination of coastal marine waters. Samples of fish liver were examined using light microscope and MICROMed digital camera. To count melanomacrophage centers (MMCs), 8-10 fields were randomly selected on each slide, captured using the camera, and readings were performed at 200X magnification. After each field of liver tissue samples have been photographed, the area of MMCs was measured (μm^2) (MICROMed digital camera software). Activity of antioxidant enzymes namely catalase (CAT) and peroxidase (PER), and aminotransferases was determined in fish liver extracts spectrophotometrically. The obtained results showed the presence of high level of MMCs, that was detected in the liver of *S. tinca* as compared with the values of *S. porcus*. Among the tested individuals in the liver of this species the greatest number of the MMCs were indicated. However, their size was 1.37% and their average size was comparable with the corresponding parameters of the *S. porcus*. It was connected with the interspecies variability of the tested fish species and specificity of their biology and ecology. CAT and PER activity in red blood cells was significantly higher in *S. porcus* (approximately 2 fold) than in *S. tinca*, while in the liver the differences between enzymes activity were less. No significant differences of aminotransferase activity were observed

in the red blood cells of two examined fish species, while in *S. porcus* liver enzymes activity was greater in 2-fold as compared with the data of the *S. tinca*. Therefore, the present results of histopathological and biochemical studies of the liver in two teleost fish species captured in coastal waters of Black Sea, demonstrated significant impact of the negative ecological conditions on fish health, caused anthropogenic pollution. Taking into account that the liver is the important metabolic organ of detoxification of xenobiotics and synthesis of main components of the blood, the examined parameters can be useful for the evaluation of its status. They could reflect the pollution level of environment and its harmful for living organisms. The data obtained are important for the environmental monitoring programs, ecological risk assessment and validation of the criteria of ecological rate of anthropogenic impact.

**Sarmurzina Z.S.¹, Zakarya K.D.¹, Bissenova G.N.¹, Abitayeva G.K.¹,
Dospayeva R.T.¹, Abzhalelov A.B.¹, Shulgau Z.T.²**

**ACUTE TOXICITY STUDY OF NEW BIOLOGICAL PREPARATION, BASED ON
LACTOBACILLUS STRAINS AND BALSAM POPLAR EXTRACT**

¹Republican Collection of Microorganisms, Astana, Kazakhstan;

²National Center for Biotechnology, Astana, Kazakhstan
sarmurzina@list.ru

The intestinal microbiota encompasses hundreds of bacterial species that constitute a relatively stable ecosystem. Alteration in the microbiota composition may arise from infections, immune defects, metabolic alterations, diet or antibiotic treatment.

We constructed a probiotic complex contains active strains of lactobacilli and balsame poplar extracts to find effective and safe biopreparat for dysbiosis.

The aim of this work was to determine the acute toxicity effect of new biological preparation on healthy rats.

Adult outbreeding rats, 20-24 g, 6-8 weeks old were obtained from National Center of Biotechnology (Astana, Kazakhstan). There were 10 rats in each group. Biopreparat was administered intragastrically. The studied preparat for the study of acute toxicity was administrated in such amounts as to determine the minimum dose which 100% of the animals die and the maximum dose which 100% will survive.

The control group of animals administered drinking water in the same volume (0.5 ml) and according to the same scheme (single dose). All animals were

sacrificed after 2 weeks of treatment. A macroscopic examination of the internal organs was carried out at the end of the experiment.

In this study, the administrations of the biopreparat at dosage 10 g / kg body weight of rats did not changes their behavior, appearance, motor activity or instant death during the period of observation.

All experiments were performed accordance with «European Convention for the Protection of Vertebrate Animals used for Experimental and Other Scientific Purposes».

The results of the present study suggest that oral administration of new biological preparation, based on lactobacillus strains and balsam poplar extract, is not toxic.

Sasanelli N.¹, Toderas I.², Iurcu-Straistaru E.², Burtseva S.³, Bivol A.², Rusu S.², Erhan D.², Byrsa M.³, Franchi M.¹

IN VITRO EFFECT OF ABAMECTIN CONCENTRATIONS AND EXPOSURE TIMES ON THE SURVIVAL OF CYST NEMATODES.

¹Institute for Sustainable Plant Protection, CNR, Bari, Italy; ²Institute of Zoology, ASM, Chisinau, R. Moldova; ³Institute of Microbiology and Biotechnology, ASM, Chisinau, R. Moldova .

nicola.sasanelli@ipsp.cnr.it

The effect of an abamectin formulation (Vertimec® EC) was tested against the cyst nematodes *Globodera pallida*, *Heterodera carotae* and *Heterodera schachtii* in an in vitro hatching test. Abamectin is a mixture of macrocyclic lactones (abamectin B1a and B1b) produced by the actinomycete *Streptomyces avermitilis*. Cysts of the nematodes were subjected to different concentrations of an aqueous solution of the abamectin formulation (0, 1.125, 2.25, 4.5, 9.0, 18.0 and 36 micro g/mL) for different exposure times. Cysts were extracted from soil samples by the Fenwick can. Batches of 50 cysts of similar size were set up and arranged according to a complete randomized block design. Each treatment was replicated three times. Untreated cysts were used as controls. Carrot root leachate, 0.3 mM zinc chloride and 0.6 mM sodium metavanadate aqueous solutions were used as natural and artificial hatching agents for *H. carotae*, *H. schachtii* and *G. pallida*, respectively. Three mL of the hatching agents were added to each batch of cysts which were incubated at 20 °C in a growth cabinet. Emerged juveniles were counted weekly. At the end of the hatching test cysts were crushed and unhatched eggs and juveniles counted. Juveniles emerging weekly were expressed as cumulative percentages of the total egg content of the

cysts (hatched + unhatched). The mortality for each treatment was calculated from percentage hatch considering the natural death in the control by the Schneider Orellis' formula. Data of percentage mortality were subjected to probit analysis to estimate values of lethal doses (LD), that is abamectin dose required for 50, 60, 70, 80, 90 and 99.9% egg mortality at each exposure time. Nematode mortality increase with the increase of abamectin concentration or at the same concentration increasing the exposure time. Abamectin LD50 of 9.9, 13.2 and 796.0 micro g/mL were calculated for an exposure time of 24 hours for *H. carotae*, *G. pallida* and *H. schachtii*, respectively. At 384 hours exposure DL50 decreased at 3.6, 2.9 and 17.6 micro g/mL for the same nematode species.

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Savitska K. O

CANDIDA ALBICANS AND PSEUDOMONAS AERUGINOSA INTERACTION DURING MULTISPECIES BIOFILM FORMATION

Odessa I. I. Mechnikov National University, Odessa, Ukraine

ksavitskaya44@gmail.com

Historically, interspecies interactions have focused on growth-inhibitory interactions, yet a variety of phenotypic outcomes other than antibiosis are possible, including alterations in developmental processes such as sporulation and biofilm formation or production of secondary metabolites.

Detecting phenotypic or developmental biomodulation between two organisms can indicate when they are communicating via small molecules, and thus can denote the presence of overlooked compounds. In other cases, signaling has been shown to occur via “repurposed” compounds – known molecules that are functioning in an unexpected manner. One exciting potential result of interspecies interactions is the induction of novel secondary metabolite production by the responding organism. Thus, examination of microbial relationships can lead to the discovery of new molecules – in some cases as the small molecule mediating the interaction, and in others as the consequent result of two microbes interacting.

To determine the microorganism interaction form during the multispecies biofilm creation co-cultivation of *C. albicans* and *P. aeruginosa* was carried out and then obtained results were compared with the sum of monospecies ones that was 100 %.

It was found that at all formation stages the cultures of a microbial multispecies association influenced each other, manifesting an antagonistic form of interactions.

The level of antagonism during the cultivation of microorganisms in the different medium under study was not the same. In the Spider medium, with ratio of the explored cultures (*C. albicans* and *P. aeruginosa*) 1:1, level of interaction already in the first day contributed to decreasing of cell number in the biofilm by almost 2 times compared to monocultures, remaining unchanged with increasing the period of cultivation.

In Saburo's medium, the level of antagonism increased over time, reaching a maximum value 48 hours after the start of cultivation with ratio of microorganisms 10:1.

Segin T.B., Hnatush S.O., Maslovska O.D.

***CHLOROBIVM LIMICOLA* IMV K-8 UNDER THE INFLUENCE CU(II) ION**

Ivan Franko National University of Lviv, Lviv, Ukraine

SeginT@ukr.net

Green photosynthetic bacteria *Chlorobium limicola* take an essential part in functioning of biocenoses, since it influences carbon and sulfur cycles because of using hydrogen sulfide as the electron donor. It results in hydrogen sulfide elimination from water environments. Often among pollutants of the environment, including man-made storages, are heavy metal compounds. Transition metal ions lead to formation of reactive oxygen species. Bacteria *C. limicola* IMV K-8, isolated from Yavorivske lake (Lviv region, Ukraine) could be successfully applied for development of biotechnologies for contaminated environments remediation from heavy metal compounds and hydrogen sulfide.

It was investigated accumulation of Cu(II) ions in *C. limicola* IMV K-8 cells after 1 hour of incubation in buffer containing copper (II) sulfate in concentration 0.05–0.50 mM. Selected concentrations of copper (II) sulfate cause reduction of biomass accumulation from 10 % to 70 %. Under the influence of all investigated concentrations of copper (II) sulfate the content of Cu(II) ions in incubation medium decreased from 12 to 16 % in comparison with sample without cells. It was shown that metal ions accumulated both on the surface of *C. limicola* IMB K-8 cells and in cells. The content of Cu(II) ions on the surface of increased with enhancing of metal salt in incubation medium. As a result of accumulation of Cu(II) ions in *C. limicola* IMB K-8 cells lipid peroxidation processes and oxidative modification of proteins were intensified. Among of the mechanisms of adaptation

of *C. limicola* IMB K-8 cells is increasing of activities of of enzymes of antioxidative system: catalase, superoxiddismutase, enzymes of glutathione system.

Among the first protection mechanisms of *C. limicola* IMV K-8 cells at the influence of Cu(II) ions also are cis/trans isomerization of monounsaturated fatty acids and formation of cyclopropane fatty acids. Maintenance of proper level of membrane fluidity provide branched chain fatty acids that possess higher resistance to the influence of reactive oxygen species.

Seidova G.M.¹, Isaeva A. F.², Muradova S.M.²

ECOLOGICAL FACTORS FORMING MYCOBIOTA OF HONEY BEES IN THE CONDITIONS OF AZERBAIJAN

¹Azerbaijan Medical University, Baku;

²Institute of Microbiology of the NAS of Azerbaijan, Baku

azmbi@mail.ru

The products of honey bees are for man not only a source of energy, also a number of minor biologically active substances. However, the consumption of these traditionally therapeutic and prophylactic products of bees now can be associated with the risk associated with their danger to humans. Since there are numerous data, that the finished products of bees can be contaminated with pathogenic microorganisms, including toxin-forming molds and yeasts. The dissemination of these products by these microorganisms is not always normalized by normative documents. In this regard, the purpose of the presented work was to study the factors of formation of mycobiota of products of honey bee and to develop a method for its regulation.

As a result of carried out studies of mycocenoses of honey bee products in ecologically different territories of Azerbaijan, were isolated and identified 72 species of micromycetes, belonging to 23 genera, the vast majority of species (52) is attributed to imperfect fungi. The mycobiota of products of honey bees are represented by the main species of the genera like as *Aspergillus*, *Cladosporium*, *Fusarium*, *Mucor*, *Penicillium*, *Rhizopus*, etc., aslo including species as *Aspergillus flavus*, *A. nidulans*, *A. versicolor*, *Penicillium cyclopium*, *Alternaria alternata*, *Fusarium oxysporum*, *F. solani* (Mart.) Sacc which are toxigenic and their mycotoxins are dangerous for humans.

Analysis of the similarity of the mycobiota of bee products to different harvesting areas revealed significant differences in practically all the products

studied, conditioned by different climatic conditions, which are manifested primarily in the species composition of fungi of the genera *Aspergillus* and *Penicillium*.

From the tested species of micromycetes isolated from the products of honey bees, 31.9% had a high toxicity with respect to the survival of the *Paramaecium caudatum* Ehren.

Besides, in the conducted researches it is established, that in ecological estimation mycobiota products of honey bees determining value have a natural - climatic zone and botanical origin of plants, as well as the type of product.

Semenets A., Finogenova M., Galkin M., Filipova T., Galkin B.

CHARACTERIZATION OF FATTY ACID PROFILES OF *PSEUDOMONAS AERUGINOSA* STRAINS WITH DIFFERENT LEVEL OF C-DI-GMP

BSEC Odessa National I.I. Mechnykov University, Odessa, Ukraine

muse-sun@ukr.net

The fatty acids are part of the structure of the cell microorganisms membrane. Cells alter the fatty acid composition of their lipids to maintain membrane fluidity with varying environmental conditions. It is essential to control the selection of a culture medium and the time and temperature of incubation before comparing fatty acid compositions with the Sherlock Libraries. Sherlock relies on qualitative (which compounds are present) and quantitative (area percentages) analysis of the fatty acid composition of organisms.

The Sherlock MIS technology uses gas chromatographic analysis of fatty acid methyl esters (GC-FAME). The Sherlock MIS Software controls a MIDI-configured Agilent Technologies GC, names the individual fatty acids in the sample and identifies the sample by comparing the GC-FAME profile to stored libraries of well-characterized strains. Post sample analysis can be used for strain tracking and microbial community profiling.

Wilde type strain *P. aeruginosa* PA01 and strains with low (PA01 pJN2133) and high (PA01 Δ wspF1) level of c-di-GMP were used as test-organisms. We identified that not all fatty acids are included in each experimental *P. aeruginosa* strains. For example, undecanoic acid is part of the cellular wall of PA01 pJN2133 at a level of 0.11%. Tridecanoic acid is part of the cellular wall of PA01 Δ wspF1 at a level of 0.10%. Palmitic acid is present in each strain at high level, however N alcohol (1-Hexadecanol) is present in only two strains PA01 (0.78%) and PA01 pJN2133 (0.61%). 12:1 3OH acid is present only in the mutant strains PA01 pJN2133 (0.79%) and PA01 Δ wspF1 (0.11%). γ -Linolenic acid present in wild-

type PA01 (0.49%) and one mutant strain PA01 pJN2133 (0.38%). However, pentadecanoic acid in its pure form is not part of the cellular wall of any strain.

Such fatty acids as, decanoic, 3-hydroxy-decanoic, dodecanoic, 2-hydroxydodecanoic, tetradecanoic, pentadecanoic iso 3-OH, hexadecanoic, heptadecanoic, cycloheptadecanoic, octadecanoic, nonadecanoic, heptadecene are present in each strain, however in different concentrations.

The fatty acid composition of different strains of *P. aeruginosa* was compared with the mobility of their cells.

Skorocho I.O., Kurdish I.K.

REDUCTION OF THE ANTIOXIDANT POTENTIAL OF THE CULTURE MEDIUM OF AZOTOBACTER VINELANDII IMV B-7076, WHEN CULTURING BACTERIA WITH SILICA NANOPARTICLES

Zabolotny Institute of Microbiology and Virology, Kyiv, Ukraine
aphalina.77@gmail.com

Over the past 10 years, nanotechnology has become one of the leading technologies in the world. There is a huge interest in the application of nanoparticles and nanomaterials in various industries. In particular, silica nanoparticles have been widely used in the biomedical and biotechnological fields.

However, when disposing of products containing nano-sized silica, its release and migration into the environment occur. A significant percentage of these nanoparticles fall into the soil. According to the literature, nano-sized silica can induce an increase in the level of reactive oxygen species (ROS) in living systems. It was interesting to investigate the effect of silicon dioxide nanoparticles on the antioxidant potential of the culture medium of *Azotobacter vinelandii* IMV B-7076 – a component of the bacterial preparation for plant growing.

At studying the effects of various doses of nano-sized silica on the antioxidant potential of the culture medium of *A. vinelandii* IMV B-7076, it was found that these nanoparticles were characterized by pronounced prooxidant effect. Thus, at the content of 0.5 g/L of silica nanoparticles in nutrient medium, all the studied parameters of the antioxidant potential of the culture medium of bacteria decreased. Among them the 2,2-diphenyl-1-picrylhydrazyl (DPPH•) radical Scavenging Activity was reduced by 21.1%, the Reducing Power – by 15.1%, Metal Chelating Activity – by 23.5%, Antioxidant Activity – by 7.7%, according to control.

As a conclusion to the studies, hat low doses of nano-sized silica either did not have a significant effect, or somewhat increased the studied parameters. At

the same time, as high concentration of silica nanoparticles, on the contrary inhibited the antioxidant and anti-radical properties of the antioxidant complex of the culture medium *A. vinelandii* IMV B-7076.

Stabnikov V.P.¹, Ivanov V.M.¹

**DEVELOPMENT OF THE NEW SCIENTIFIC DISCIPLINES OF
CONSTRUCTION MICROBIOLOGY AND CONSTRUCTION BIOTECHNOLOGY**

¹National University of Food Technologies, Kyiv, Ukraine

vstabnikov1@gmail.com

The new scientific and engineering disciplines, Construction Microbiology and Construction Biotechnology, are developing exponentially during the last decade. The major directions of Construction Microbiology are selection of microorganisms participating in construction bioprocesses and study of their ecology, physiology, and molecular biology. The major directions of Construction Biotechnology are development of the microbially-mediated construction biotechnologies for the use in construction industry. The products of construction biotechnologies are low cost, sustainable, and environmentally friendly microbial biocements and biogroups for the construction ground improvement. The microbial polysaccharides are used as the admixtures for cements and mortars. Microbially produced biodegradable bioplastics can be used for the temporarily constructions. The bioagents that are used in construction biotechnologies are either selected pure microorganisms, however in some cases enrichment cultures or activated indigenous microorganisms of soil also can be used. The major physiological groups of bacteria that can be used in construction processes are halotolerant urease-producing, iron-reducing, denitrifying, and acetate-oxidizing bacteria. The applications of microorganisms in the construction processes are soil bioaggregation, biocementation and bioclogging of porous materials and fractured rocks, biodesaturation of unstable construction ground, immobilization of soil pollutants, dust and soil erosion control. The biotechnologically produced construction materials and the microbially-mediated construction technologies have a lot of advantages in comparison with the conventional construction materials and processes. Proper practical implementations of construction biotechnologies could give significant economic, environmental, and social benefits, see for example.

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Stetska V., Holota Y., Bogomolova K., Dovbunchuk T., Serhiychuk T., Tolstanova G.

LONG-TERM EFFECTS OF CEFTRIAXONE AND COMPOSITION AMPICILLIN WITH METRONIDAZOLE ON COLONIC MICROBIOTA IN RATS

Taras Shevchenko National University of Kyiv, Ukraine

victoria_stetska@ukr.net

Antibiotic-associated dysbiosis is detected in 5-35% of patients, during or immediately after antibiotic withdrawal. The degree of dysbiotic changes depends on the specific type of antibiotic, the dose and the term of its use. In our previous study we found progressive, long-term changes in colonic microbiota (CM) composition after 14-days treatment with antibiotic ceftriaxone (Cf). The aim of the present study was to compare the changes in CM after administration of Cf or compositions of ampicillin and metronidazole (Amp/Met).

Methods. Studies was done on male Wistar rats (170-200g, n=22). Cf (300mg/kg, PJSC "Kyivmedpreparat") was injected i.m for 14 days. Composition of Amp (75mg/kg, Kyivmedpreparat PJSC) and Met (50mg/kg, Ph.C. "Zdorovya") was injected once a day for 3 days per.os. of the composition of faecal and mucosa-associated microbiota was detected bacteriologically on the 1st day, in 2 and 8 weeks after antibiotic withdrawal.

Results. Immediately after antibiotics withdrawal, more pronounced microbiotic changes were observed after in Amp/Met treated rats. There was increase the number of conditionally pathogenic enterobacteria (CPE) (from $1g\ 0.75\pm1.33$ to $1g\ 4.00\pm0.00$ CFU/g), appeared lac(-) *E.coli* ($1g\ 4.05\pm4.07$ CFU/g) and hemolytic forms ($1g\ 3.85\pm3.91$ CFU/g). The number of lac(+) *E.coli* and bacteria of the *Clostridium* genus increased 1-2 fold. On the first day after Cf withdrawal lac(+) *E.coli* wasn't detected, and appeared *Staphylococcus spp.* ($1g\ 4.68\pm0.58$ CFU/g). Two weeks after Amp/Met withdrawal, the gradual normalization of the microbiota was observed. While, changes in microbiota composition in Cf-treated rats were still observed. In 8 weeks after Amp/Met withdrawal, the normalization of the CM was noted, while dysbiotic changes in Cf-treated rats further progressed. The number of *Bifidobacterium* and *Lactobacillus* were decrease in 1-2 fold. The increased number of *Clostridium* (to 3.18 ± 0.48 CFU/g), lac(-)&lac(+) *E.coli* 2-fold and CPE (from $1g\ 1.08\pm0.44$ to $1g\ 6.02\pm0.68$ CFU/g) were detected.

Conclusions. For modeling of acute dysbiosis, which develops immediately after antibiotic withdrawal, the model with Amp/Met is more adequate. For modeling the long-term effects of antibiotics, it's recommended to use models with Cf.

Stetska V., Vypovska O., Dovbunchuk T., Serhiychuk T., Tolstanova G.
FECAL MICROBIOTA IN RATS WITH 6-OHDA INDUCED PARKINSON'S
DISEASE

Taras Shevchenko National University of Kyiv, Ukraine..

victoria_stetska@ukr.net

Parkinson's disease is the second in the frequency of occurrence chronic, slowly progressive neurodegenerative disease after Alzheimer's disease that characterized by the loss of dopaminergic neurons in substantia nigra, which etiology remains unclear. Researches of last years indicate to a possible relationship between changes in the intestinal microbiota and the development of inflammation on the periphery as the trigger factors of neuroinflammation and respectively neurodegeneration. But all the information remains at the level of assumptions and requires experimental proof. The aim of this work was to analyze the changes in the colon's microbiota in 6-OHDA induced Parkinson's disease in rats.

Methods. The study was done on male Wistar rats (n=10, 230-300g). Parkinsonism was modeled by one-sided destruction of the dopaminergic neurons of a compact part of the substantia nigra, by injection 12 µg selective neurotoxin 6-OHDA in the left lateral upward proximal bundle. To determine the microbiota of rats feces (1g) were homogenized in 9ml of sterile 0.5% sodium chloride solution. From the obtained homogenate were prepared ten times dilution (10⁻¹-10⁻⁸). The quantitative and qualitative changes of microbiota composition were determined by sowing dilutions on differential diagnostic media with selective properties: Bifidobacterium Agar; MRS agar; Endo; Iron Sulfite Agar; Mannitol Salt Agar, Simmons Citrate Agar; 5% blood agar. Cultivated 24-48 hours at 37°C. Identification of isolated microorganisms was carried out according to morphological and tinctorial characteristics.

Results. As part of the luminal microbiota of rats, within two months from the start of modeling of parkinsonism, significant changes were detected only for *E. coli*. So the amount of lac(+) *E.coli* increased from 1.5 months to 2 orders (from lg4.65±0.80 to lg6.08±0.70 CFU/g (1.5 months) and from lg4.39±0.55 to lg6.24±1.26 CFU/g (2 months). At the same time, the amount of lac(-) *E. coli* decreased by 2-3 orders.

Conclusions. The tendency to increase the number of Gram-negative enteric bacilli, which have lipopolysaccharide in their cell wall are capable of inducing local synthesis of alpha synuklein that may promote neurodegenerative process.

**Stetska V.¹, Gonchar S.¹, Sholokh A.¹, Dovbunchuk T.¹, Issaeva O.²,
Semenikhina M.², Nikolaienko O.², Serhiychuk T.¹, Tolstanova G.¹**

EFFECT OF PILOCARPINE INDUCED EPILEPSY ON COLON'S MICROBIOTA IN RATS

¹Taras Shevchenko National University of Kyiv, Ukraine;

²Bogomolets Institute of Physiology of the NASU, Kyiv, Ukraine

victoria_stetska@ukr.net

Epilepsy (Ep.) is a heterogeneous disorder with comprehensive etiology and needs for few therapeutic approaches. One of the promising horizons of the neuroscience complex is the microbiota-brain axis, explained by the ability of microorganisms to synthesize neurotransmitters making effect on the nervous system. At the same time, there are some reports about the possibility of therapy of neurodegenerative diseases (ND) by correction of the intestinal microbiota by probiotic microorganisms. The rational use of probiotics and understanding the relationships between microbiota and the brain need knowledge about the effect of the ND on the colon's microbiota in the dynamics of disease development. The aim of this work was to investigate the changes in the mucosa-associated microbiota (MAM) of the colon and translocation of bacteria in the blood of the portal vein in rats with pilocarpine-induced Ep.

Methods. The study was done on male Wistar rats (170-200g, n=22) with induced Ep. by pilocarpine (40 mg/kg) injection. Autopsy was done in 1 (acute) or 14 (chronic) days after epileptic seizures development. The number of microorganisms of the colonic MAM and blood of the portal vein were determined bacteriologically by sowing 10-fold the dilutions of the investigated material on the selective differential media.

Results. Changes in microbiota composition were recorded only at acute stage of Ep. There were a significant increase number of *E.coli* (from $\lg 3.04 \pm 1.95$ to $\lg 6.21 \pm 1.68$ CFU/g) and *Staphylococcus* (from $\lg 0.43 \pm 1.05$ to $\lg 3.02 \pm 2.19$ CFU/g, $p < 0.05$) in colonic mucosa. In animals that were administered pilocarpine, but weren't recorded epileptic seizures (control group), all indices were within the range for intact animals. Pilocarpine-induced Ep. was associated with increase in the bacteria translocation of portal vein both at the acute and chronic stages of disease.

Conclusions. The significance of *E.coli* growth in the MAM can be considered from two points: on the one hand, it's an increase in the lipopolysaccharide pool, which may be a factor in ND and on the other hand in *E.coli* a protein was found that capable of inhibiting the synthesis of amyloid that plays a leading role in the emergence of ND.

Strezeva L. M., Dzhura M.S., Galkin M.B., Filippova T.O.

**ACTIVITY OF 2-HEPTYL-3-HIDROXY-4-QUINOLON SYNTHETIC
DERIVATIVES ON GROWTH AND BIOFILM FORMATION OF *PSEUDOMONAS
AERUGINOSA* STRAINS WITH DIFFERENT LEVEL OF C-DI-GMP**

Odessa National I.I. Mechnikov University

kiko.tsuki@gmail.com

The aim of this work was to investigate the effects of PQS derivatives on the planktonic culture growth and biofilm formation of *P. aeruginosa* strains. We used PQS and its synthetic derivatives of 2-propyl-3-hydroxy-4-quinolone (C₃Q) and 2-pentyl-3-hydroxy-4-quinolone (C₅Q) which were synthesized at the Biotechnological Research Center of ONU. Due to the fact that the biofilm formation is under control of the intracellular level of the secondary messenger of cyclo-di-GMP, the following strains were used in the work: *P. aeruginosa* PA01 (wild strain strain), *P. aeruginosa* ΔwspF1 (strain with an elevated level of c-di-GMP and increased ability to form a biofilm) and *P. aeruginosa* pJN2133 (a strain with a reduced level of c-di-GMP and a reduced ability to form a biofilm).

Obtained results indicate that in concentrations of 20, 40 and 80 μM, their effects on the investigated parameters were correlated with the intracellular level of c-di-GMP. Thus, all compounds showed moderate ability to reduce the number of cells in a planktonic culture, except for C₅Q - which, at a concentration of 80 μM significantly inhibited its growth by 50-70%. Studded compounds show an ability to inhibit biofilm formation process *P.aeruginosa* PA01 and *P. aeruginosa* ΔwspF1. In this case, moderate inhibition was observed within the first strain, but in *P. aeruginosa* ΔwspF1 it was more significant- almost two times in the presence of 80 μM C₅Q compare to control. In contrast, the ability to form a biofilm in *P. aeruginosa* pJN2133 significantly increased in the presence of descovored substances. In presence of investigated compounds at concentration of 80 μM PQS, the biofilm mass was 315% compared to control; in presence of C₃Q - 276% and C₃Q - 256% respectively.

Obtained results may indicate that PQS and its synthetic derivatives can affect not only the functioning of the quorum sensing system, but also the exchange of c-di-GMP, which makes them perspective tools for regulating the work of both systems and is promising for further research.

Tkachuk N.V.¹, Zelena L.B.²

**IDENTIFICATION OF IRON-REDUCING BACTERIA ISOLATED FROM SOIL
FERROSPHERE**

¹National University «Chernihiv collegium» named after Taras Shevchenko, Chernihiv, Ukraine;

²Zabolotny Insitute of Microbiology and Virology, NAS of Ukraine, Kyiv, Ukraine
nataliia.smykun@gmail.com

In microbial-induced corrosion processes, conditions for the development of sulfate-reducing bacteria are created by heterotrophs, in particular ammonifying and iron-reducing bacteria.

The aim of the present research was to isolate and identify iron-reducing bacteria from the soil ferrosphere using microbiological, physiological-biochemical and molecular genetic methods, to analyze their ability to produce some corrosive dangerous metabolites.

Iron-reducing bacteria were isolated from the soil ferrosphere and selected on FWA-Fe (III) citrate medium by Koch's method. Analysis of a complex of microbiological, physiological and biochemical characteristics, 16S rRNA gene sequencing and phylogenetic analysis were performed using conventional methods and techniques.

ChNPU ZVB1 strain was isolated and selected as one with iron-reducing and ammonifying properties and the ability to form hydrogen sulfide. According to the Bergey's Manual of Systematic Bacteriology the strain was identified as *Bacillus* sp. on the basis of the complex of microbiological, physiological and biochemical properties. For further identification of the strain 16S rRNA gene amplification followed by sequencing of the PCR product was carried out. The resulting partial sequence was compared with those deposited in the GenBank database. The result of the analysis showed 99% similarity to the different species of *Fictibacillus* and *Bacillus* genera. 16S rRNA gene nucleotide sequence of the ChNPU ZVB1 strain was deposited in the GenBank database as *Fictibacillus* sp. KX349222. To define the phylogenetic position the dendrogram of relationship between various *Bacillus* and *Fictibacillus* species was constructed. There were two large clusters: the first was composed of species belonging to *Bacillus* genus, and the other combined species of *Fictibacillus* genus, including the *Fictibacillus* sp. strain ChNPU ZVB1. The closest neighbors to the *Fictibacillus* sp. strain ChNPU ZVB1 were *F. phosphorivorans* and *F. nanhaiensis* type strains. Thus, based on the obtained results the ChNPU ZVB1 strain belongs to the *Fictibacillus* genus and its species identification require additional molecular-genetic analysis.

Toderas I.¹, Iurcu E.^{1,4}, Burtseva S.², Birsa M.², Bivol A.¹, Rusu S.¹, Caldari V.¹, Sasanelli N.³

STIMULATION OF SUGAR BEET SEEDS GERMINATION BY PRODUCTS OF STREPTOMYCETES METABOLISM AS BIOTECHNOLOGICAL METHOD FOR TO PROTECT AGAINST CYST-NEMATODES OF GENUS *HETERODERA*

¹Institute of Zoology, A.S.M., Chisinau, Republic of Moldova;

²Institute of Microbiology and Biotechnology, A.S.M., Chisinau, Republic of Moldova;

³Institute for Sustainable Plant Protection, C.N.R., Bari, Italy;

⁴Tiraspol State University, Republic of Moldova.

iurcuelena@mail.ru

Sugar beet is an important agro-industrial crop fundamental in the agroecosystems of the northern part of the Republic of Moldova. The phytoparasitic nematodes of the genus *Heterodera* can seriously damaged the crop. Therefore, researches to reduce the impact of these pests by phytonutrient complexes are needed. The adaptation of biotechnological measures as germ-type phytostimulation is an interesting method for an ecological protection. Recently, some researches have been explored in the germination process as a means of accelerating the formation of the root system, accompanied by lignification processes as barriers to hamper the parasitic impact with invasive cyst-forming larvae of the genus *Heterodera*. Investigations to obtain exometabolites of strains of isolated streptomyces from soils of the Republic of Moldova were carried out within the National Collection of Non-pathogenic Microorganism of the Institute of Microbiology and Biotechnology (ASM, R. Moldova). Exometabolites were obtained by streptomycetes strains cultivation on liquid nutritive medium M-I and culture broth was separated from biomass by centrifugation.

Sugar beet variety, approved for cultivation in the Republic of Moldova, was tested for the sprouting process with 8 different exometabolite solutions at 0.5% and 1.0 % concentration. Growth dynamics and formation of sugar beet roots were observed at 24 hours and 14 days after treatments. Exametabolites of strains 9, 33, 47 and 49 were effective to stimulate and improve root growth from 24.7 to 34.2%, compared to the untreated control, resulting in a recommended biotech remedies with potential opportunities for registration and patenting as method to limit the phytoparasitic impact of invasive parasite complexes on sugar beet in R. Moldova.

The investigations were carried out with the financial support of the bilateral Moldova - Italy (ASM/CNR, years 2015-2016) and STCU – 6233 projects and of

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Tokovenko I.P.

PECULIARITIES OF AMINO ACIDS METABOLISM OF PHYTOPLASMAS

Zabolotny Institute of Microbiology and Virology, Kyiv, Ukraine

tira@bigmir.net

Phytoplasmas are a special group of pathogens of plants with a minimum genome size. At present more than 70 diseases of plants caused by phytoplasmas are known. The pathogenicity of phytoplasmas in infected plants is very high. Metabolic activity of phytoplasmas is limited by their biosynthetic possibilities, which are caused by the minimum amount of genetic information for cells of these pathogens and, accordingly, the minimal number of metabolic pathways, which ultimately leads to a significant dependence on the presence of nutrient substrates in the nutrient medium. Consequently, in order to maintain their existence, phytoplasmas need different nutrient substrates that they could take outside - either from the nutrient medium in which they are grown or from the host cells in which they are inhabit. Among the nutrient substrates required for these pathogens to exist, amino acids, as the main sources of energy, nitrogen and carbon, take on the main place. What spectrum of amino acids is necessary for these pathogens to support their growth and development? Consider this question. The conducted researches have established that phytoplasmas are capable to consume a considerable quantity of amino acids. Thus, all investigated phytoplasmas assimilated asparagine, glutamine, threonine, histidine and proline. In addition, majority of strains have assimilated phenylalanine, methionine, glutamate, lysine and alanine. Somewhat unexpected was the worse assimilation of tyrosine by phytoplasmas compared with phenylalanine. More than half of the examined strains included in their metabolism of cysteine and leucine. It is important to note that a significant amount of phytoplasmas had the ability to decompose arginine. A low ability to assimilate isoleucine and tryptophan was noted, which may indicate the ability of these pathogens to synthesize these compounds. Thus, it has been found that for metabolic needs of phytoplasmas necessary such amino acids as asparagine, glutamine, threonine, histidine, proline, phenylalanine, methionine, glutamate and lysine. Such a need in amino acid testifies to insufficient own biosynthetic capabilities and indicates to their parasitism.

Trofimenko Yu. Yu., Tretiakov M.S. , Nazarchuk O.A.

**MICROBIAL COLONIZATION OF TRACHEAL TUBES IN PATIENTS OF
INTENSIVE CARE UNIT**

National Pirogov Memorial Medical University, Vinnytsya Ministry of Health of Ukraine,
Vinnytsya, Ukraine

yuliya-trofimenko@ukr.net

The implantation of polymeric medical devices inside the human body is accompanied with microbial colonization. The prolonged stay of endotracheal intubation tubes (EITs) in the respiratory tract creates preconditions for formation of unusual for healthy mucous microbiota in the respiratory tract of human.

The aim of the research was to study the characteristics and biological properties of species composition of bacteria, isolated from the surface of endotracheal intubation tubes in patients of the Department of Intensive Care Unit (ICU).

Materials and methods. A total of 36 EITs, introduced not less than 4 days into the respiratory tract of patients in the intensive care unit, have been studied. Determination of the nature of microbial colonization EITs were carried out by cropping from the surface of the tube segment while rolling through the plane of a dense nutrient medium. In the research we isolated 85 clinical strains of microorganisms.

Results and discussion. The received data demonstrated that in patients, who had been treated in the ICU, there were predominantly found non-fermenting Gram-negative bacteria on the surfaces of EITs. *Acinetobacter* (60,4%) were identified as leading ones. At the same time, representatives of the specified species were allocated as part of microbial associations in 52,1% of cases, and only 8,3% were found in monoculture. Among other non-fermenting rods *Pseudomonas spp.* (16,7 %) and *Stenotrophomonas spp.* (8,3 %) were isolated in microbial associations. The amount of *Enterobacteriaceae* representatives was also huge in the composition of microbial associations. Microbes of the genus *Klebsiella* and *Enterobacter* were isolated the same frequently (16,7%). Bacteria of the genus *Escherichia* were isolated from the EITs surfaces of 8,3% of patients. Among the Gram-positive bacteria, there were identified *Enterococcus spp.* (29,2 %), *Staphylococcus spp.* (8,3 %).

Conclusion. Opportunistic microorganisms take part in colonization EITs surfaces introduced in respiratory tract of patients of ICU, among them such as non-fermenting Gram-negative bacteria, *Enterobacteria*, *Staphylococci* and *Enterococci* are of high specific gravity.

**Tugay A.V.¹, Tugay T.I.^{1,2}, Zheltonozhsky V.A.³, Zheltonozhskaya M.V.³,
Sadovnikov L.V.³, Bulanchuk Yu.M.¹**

**CHANGES IN THE PROOXIDANT AND ANTIOXIDANT SYSTEMS IN THE
FOUR POST-GENERATION GENERATIONS OF *ASPERGILLUS VERSICOLOR*
IN THE EFFECTS OF IRRADIATION**

¹D.K. Zabolotny Institute of Microbiology and Virology of the NASU;

²Open International University of Human Development 'Ukraine';

³Institute for Nuclear Research

andre.07111982@gmail.com

The study of the effects of chronic ionizing radiation on microbiota is important because it is continuous and active in the composition of biogeocoenosis and plays a significant role in the transfer of various nutrients and some trace elements in the soil, including radionuclides. For micromycetes there is a rapid change in generations, which makes them a convenient model for studying the effects of chronic irradiation in a number of generations, the establishment of direct and long-term effects of chronic ionizing radiation.

To study long-range effects, a combined laboratory-field approach was used.

The study of the functioning of pro- and antioxidant systems in postradiation generations of *Aspergillus versicolor* strains isolated from background areas and not showing radio adaptive properties and strains isolated from the territory of the Exclusion Zone, which are radio adaptive have been studied.

The active accumulation of diene conjugates in postradiation generations of the strain that does not exhibit radio adaptive properties and accumulation at the control level in postradiation generations of the strain with radio adaptive properties is revealed. An increase in the content of malonic dialdehyde was found only in the first generation of strains that did not exhibit radio adaptive properties and at the control level in the strain with radio adaptive properties. The active accumulation of trienium conjugates (8 times) was found only in the third generation of strains that did not exhibit adaptive properties of the radio, and in the strain with adaptive properties of the radio it was detected in the second and third generations. An increase in superoxide dismutase activity in postgraduation generations of a strain that did not exhibit radio adaptive properties and changes at the control level in the strain with radio adaptive properties was found. The changes in the control level in postradiation generation strains that showed no radio adaptive properties and increased in the third and fourth in the strain with radio adaptive properties were detected in catalase activity.

Tymoshok N.O., Gulaeva G. B., Spivak M.Ya

NANO-SELENIUM EFFICACY ON BIOLOGICAL PROPERTIES OF *BACILLUS SUBTILIS*

Zabolotny Institute of Microbiology and Virology of the National Academy of Sciences of Ukraine, Kyiv

N.Tymoshok@ukr.net

Influence of nano-selenium (SeNP) on the rate of producing extracellular biological active substance and biomass growth have been studied on *Bacillus subtilis* IMV B-7392. At SeNP-treated bacteria in the range of concentrations from 2×10^{-2} to 2×10^{-8} mg/ml was without changes in the culture and morphological characteristics of bacterial cells. Increasing the biomass growth and physiological responses of *Bacillus subtilis* was observed when the SeNP concentration was 2×10^{-4} mg /ml in modified Gauze medium. The holding in the 0.15M NaCl of this culture (1000 cells / ml) separately and in combination with SeNPs at optimum concentration for 24 hours, allowed to increase the yield of biologically active substances significantly. The protein contents in samples was analyzed spectrophotometric measurements at λ 235 -280 nm. Proteins of extracellular biological active substances of *Bacillus subtilis* showed lectin activity, in addition, stimulated the growth and development of cereal plants, with influencing adaptive metabolic processes of plants, as well as maintaining normal functional condition of photosynthetic apparatus.

Vorobieva N.G., Yumyna J.M., Hrytsev O.A.

DETECTION OF QUARANTINE AND REGULATED NON-QUARANTINE BACTERIAL DISEASES IN SEED POTATO MATERIAL

Taras Shevchenko National University of Kyiv, Ukraine

tyska16.09@ukr.net

Major bacterial diseases like ring rot, bacterial wilt or brown rot cause considerable loss to potato production. *Ralstonia solanacearum* is the causal agent of the disease known as potato brown rot or bacterial wilt. The regulated non-quarantine organism *Clavibacter michiganensis* subsp. *sepedonicus* (Cms), the causative agent of bacterial ringrot in potato, poses a constant threat to potato growers in Europe. Spread of the pathogens worldwide has been associated with its dissemination in latently infected seed material.

The purpose of this study was to detect quarantine and subquarantine bacterial pathogens in seed potato material using the DAS ELISA method.

Sampling of potatoes was carried out in February and March 2016 in accordance with the requirements of DSTU 4014-2001. The work was done under conditions of the 2nd class of protection laminar boxing taking all aseptic measures.

The samples under study were placed in homogenization bags and diluted in a bulk ratio of 1:20 in buffer for conjugation. Homogenization of the samples was carried out using an automatic homogenizer HOMEX 6 (Bioreba, AG Switzerland).

The presence of pathogens was determined by the enzyme-binding immunosorbent method (double antibody sandwich) DAS ELISA using commercial test systems LEOWE® Standard Complete Kit (Germany) according to the manufacturer's instructions. An antibody incubation was performed at a temperature of 37°C in a thermosheet for microplates PST-60HL-4 (Biosan, Latvia), followed by 4-time automatic washing with the help of PW-40 (Bio-Rad, USA).

Reading the results was carried out in a microplate photometer SUNRISE (TECAN Austria GmbH, Austria) at a wave length of 405 nm. The Magellan V.7.1 program was applied to process the results. The morbidity level (%), or the affection of tubers (%) was determined by the formula: $P = A \cdot B \cdot 100 / A$, where P is the morbidity level, or morbidity; A is a total number of tubers; B is the number of healthy tubers.

The morbidity level of seed potato for 4 batches of the ring rot causative agent (*Clavibacter michiganensis* subsp. *sepedonicum*) made 25%; for the bacterial wilt (*Ralstonia solanacearum*) it was 28%.

Voronkova O.S.

ABILITY TO BIOFILM FORMATION OF STAPHYLOCOCCAL STRAINS, ISOLATED FROM DIFFERENT BIOTOPES OF HUMAN BODY

Oles Honchar Dnipro National University, Dnipro, Ukraine

voronkova_olga@i.ua

Analysis of the spreading of the ability to form a biofilm among the strains of staphylococci, isolated from the reproductive tract of women, upper respiratory tract, skin, gastrointestinal tract (intestine and oral cavity) of healthy individuals and persons with pathological manifestations, and among the strains isolated from abiotic surfaces, showed that the frequency of detection of staphylococci in these biotopes during pathological processes is not less than 75%. Among staphylococcal strains *Staphylococcus aureus* (46.3%) and *Staphylococcus*

epidermidis (38.3%) were the dominant species that depends on the biotope. The frequency of detection of film-forming strains also varied depending on the biotope / surface and ranged from 35% to 100%. The percentage of detection of film-forming strains from healthy individuals was significantly lower: the index was close to 15%, that could be considered as evidence of the importance of the formation of biofilms in the development of complications. It should be noted that the rate of detection of film-formation was greater among *S. epidermidis* strains, of which at least 50% were able to form a biofilm, while for *S. aureus*, this indicator began with 35%. In addition, it should be noted that the ability to film-formation was predominantly determined in strains isolated during chronic or prolonged lesions, while during the acute forms of lesions the frequency of detection of such strains was minimal. For example, during dysbiosis of the gastrointestinal tract the part of film-forming strains was over 79%, while during poisoning it was only 23.8%.

It should be noted that for film-forming strains isolated from persons without manifestation of a pathological process, the rates of adhesion were lower. Among the film-forming strains there were no any non-adhesive strains, and high-adhesion strains (over 75-80%) prevailed, while among the non-film-forming strains the predominant groups were medium- and low-adhesion strains. Comparison of the properties of strains of *S. aureus* and *S. epidermidis* showed that the strains of epidermal staphylococci in most cases differed by higher rates of adhesion.

Vozniuk S.V.¹, Tytova L.V.¹, Panchenco A.M.², Iutynska G.O.¹

BIODIVERSITY OF BACTERIA OF THE RHIZOBIALES ORDER IN THE SOY RHIZOSPHERE MICROBIOME UNDER APPLICATION OF FUNGICIDES AND COMPLEX INOCULATION

¹D.K.Zabolotny Institute of Microbiology and Virology of the NAS of Ukraine, Kyiv, Ukraine;

²Taras Shevchenko National University of Kyiv ESC "Institute of Biology and Medicine", Kyiv, Ukraine

vozsvet@gmail.com

Soil microbiome biodiversity preservation is important for agroecosystems stabilization, their stress resistance and productivity.

Soy is a strategic legume crop of the world agriculture. Lately, the phytopathogenic fungi accumulate in soybean crops. The fungicides are used for the mycoses control, but their influence on the rhizosphere microbiocenosis is insufficiently studied.

The subject of this study was the bacteria of Rhizobiales order of Proteobacteria phylum, which include agronomically useful microorganisms. There are bacteria of bioformulation among of them. The soy seeds were treated by fungicides Maxim Star or Kinto duo, followed by bioformulation Ecovital inoculation. Ecovital consist of *Bradyrhizobium japonicum* and *Bacillus megaterium* strains. The total DNA of rhizosphere soil of Annushka cultivar soy was studied using the high-performance pyrosequencing method.

In the soy rhizosphere microbiome 21 phylums were identified, one of which belonged to *Archaea*, *Acidobacteria*, *Actinobacteria*, *Bacteroidetes*, *Firmicutes* and *Proteobacteria* phylums were dominant. The representation of all phylums, except *Actinobacteria*, increased in variants with fungicides seeds treatment and inoculation in 1.3-4 times in comparison with the control variant. The share of *Bradyrhizobiaceae*, *Rhizobiaceae*, *Methylobacteriaceae*, *Phyllobacteriaceae*, *Xanthobacteraceae* and *Hyphomicrobiaceae* families increased in the inoculated plants rhizosphere. Representatives of *Hyphomicrobiaceae* and *Xanthobacteraceae* families were found only in experimental variants. In the soy rhizosphere for the first time it was detected *Balneimonas* and *Bosea* genera representatives (family *Bradyrhizobiaceae*), which previously were found in typical chernozem of wheat and sugar beet rhizosphere. Bacteria of *Bosea* genus were found in all experimental variants, but *Balneimonas* were detected only in the variant with the fungicide Maxim Star and Ecovital combined application. In the same variant, we found bacteria of *Kaistia* genus (*Rhizobiaceae* family).

Thus, the combined use of fungicides and inoculation contributed to the increase of *Rhizobiales* order representatives diversity in the soy rhizosphere microbiome.

Yablonska O.V., Savchuk O.V., Mekh N.Ya.

FOOD MICROFLORA AT THE MEAT DURING STORED

National University of Life and Environmental Sciences of Ukraine, Kyiv, Ukraine

yablonska.oksana@gmail.com

Microbiological expertise of meat products must be started at the industrial level, at the sanitaria-epidemiological and veterinary medicine lab at the market. Nota Bene: raw contaminated meat does not change.

During the research we used analytical, statistical and microbiological methods. We conducted analytical studies of microbiological data over the past 10 years, which are sent to the Scientific Research Institute for Laboratory Research and the veterinary and sanitary examination.

Samples of meat products were vacuum sampled for laboratory testing.

The our research showed that in general in Ukraine meat products are as clean as possible. The greatest contamination with microorganisms was determined in the north, east and several south-western regions (for ethical reasons we do not call specific areas).

All samples from the products of some eastern regions contained *Salmonella*, *Escherichia coli*, *Proteus*, and also cocci and anaerobes. In the products of some northern and southern regions, only *Salmonella* was identified, in particular in the north in beef 132 strains, in poultry — 30 strains, and in the south — in the beef 180 strains of *Salmonella*, and in chicken — 13 strains. The same products in the south-western regions were contaminated with *Salmonella*. Thus, in the beef 23 strains were detected, in the poultry — 73 strains. Contamination with *Salmonella*, cocci and *Escherichia* of this type of meat was observed in all these areas.

The results of our laboratory studies are similar to the statistical data of laboratories. Most of the products tested were pure from a microbiological point of view.

On the surface of 20 per cent of samples of meat products that are sold in vacuum packs, we determined mainly saprophytic and opportunistic microorganisms (*M. citreus*, *Staph. epidermidis* and *E. coli*).

The microbial background of 5 per cent of samples of meat products consisted of a conditionally pathogenic & pathogenic microbes (*Staph. epidermidis*, *Staph. aureus*, *Streptococcus pyogenes*, *Bac. subtilis*, *E. coli*, *Ps. aeruginosa* and *S. marcescens*), the amount of which varied depending on the storage conditions, its duration and temperature.

Yablonska O.V.¹, Kasper V.S.²

EFFECTIVENESS OF THE USE SYNBIOTIC LACTIALE AFTER ANTIBIOTIC THERAPY IN DOGS

¹National University of Life and Environmental Sciences of Ukraine, Kyiv, Ukraine;

²Odessa State Agrarian University, Odessa, Ukraine

vab321bav@gmail.com

Currently, probiotics are widely used in the treatment of widespread diseases of different etiologies in Ukraine.

However, there is space for the use of synbiotics, because they are rarely used because of the lack of a clear understanding of the differences in probiotics from synbiotics. In general, the appointment of probiotics and synbiotics

physicians are not well-considered, considering these drugs ineffective and poor-quality. But in fact, such drugs have a great influence on the metabolism of the body. Some researchers consider our internal ecosystem as a separate body, which is important for the whole body as well as other body organs. Intestines contain more microorganisms than cells that make up the human body or animal.

Dr. Roy D. Slitter of the Irish Institute of Cork believes that the true role of bacterial populations living in the body is too low. According to his data in the intestine contains almost two kilograms of bacteria. Dr. Slytor also stresses the constant use of probiotic drugs, which ensures their strengthening on the intestines and the organization of colonies of beneficial bacteria. Especially when the body is weakened after antibiotic therapy.

The most common synbiotics include: Multilac, Duopic, Lacial, Bifitin, Bifidobac, Bifidumbacterin 1000, Normospectrum, Enterronorm Detox.

The analysis of literature data showed that the appointment of synbiotics for the prevention of intestinal disorders and bacterial infections in dogs and cats is more rational than the use of probiotics.

According to the results of the investigation in two groups of dogs, the composition of the intestinal microflora was determined. Prior to treatment with Lactiale in the first group, there were 0.6% bifidobacteria and 0.4% lactobacilli in the first group, and in the second 0.5% bifidobacterium and 0.5% lactobacilli. After the treatment in the experimental group, the composition of the microflora became normal (60% bifidobacteria, 17% lactobacillus), and in the control group, the rates were lower than normal (35% bifidobacteria, 8% lactobacillus).

Consequently, the appointment of synbiotics - is a more rational and effective method.

Yaroshko O., Kuchuk M.

OBTAINING OF TRANSGENIC ROOTS OF *AMARANTHUS CAUDATUS* L. AFTER CO-CULTIVATION WITH *AGROBACTERIUM RHIZOGENES* A4

Institute of Cell Biology and Genetic Engineering NAS of Ukraine, Kyiv, Ukraine
90tigeryaroshko90@gmail.com

Amaranth becomes popular due to its rich biochemical composition, it is promisingly to improve it with biotechnological methods.

The objects of investigation were *Amaranthus caudatus* L. cv. *Karmin* and wild strain of *Agrobacterium rhizogenes* A4.

The purpose of the work was to optimize the conditions for co-cultivation of cv. *Karmin* with *A. rhizogenes* and to obtain transgenic roots for this cultivar.

Seeds germinated on sterile medium Murasige and Skuga (MS₃₀) with 30 g/l sucrose. For transformation were used hypocotyl segments of 15-day-old seedlings, leaves and internode parts of 2-month-old plants. Transformation was carried out by co-cultivation of explants with *A. rhizogenes*.

First, bacterium was sowing on liquid LB medium with acetosiringone for 24 h. Next, bacterium was centrifuging during 12 min, 5000 rpm. and resuspended into liquid medium ½MS₁₅. In this medium explants were soaked for 0 (control), 5, 10, 15, 20, 25, 60, 90 minutes.

Next step was transfer of explants on the medium ½MS₁₅ without antibiotics. Cultivation on this medium continued for 0 (for this variant, after pre-cultivation explants were transferred immediately on the medium ½MS₁₅ with 500 mg/l of cefotaxime), 1, 2, 3, 4 days.

Then, the explants were transferred on the medium ½MS₁₅ with 500 mg/l cefotaxime. Every 2 weeks explants were transferred on the ½MS₁₅ medium with 400 mg/l, 300 mg/l, 200 mg/l of cefotaxime. In the last transfer used ½MS₁₅ medium without cefotaxime. The following sub-cultivation for control samples didn't conduct, because after 15 days explants died out.

Growth of "hairy roots" on the explants observed after 25 days after transformation. The positive results of obtaining transgenic roots for 2 samples were confirmed with PCR analysis.

So, it was established that the most effective conditions were co-cultivation of 2-month-old internode explants during 1 day with *A. rhizogenes*. The most effective pre-cultivation time was 90 minutes.

Yunosheva O.P., Ellanska N.E.

**ANTIMICROBIAL PROPERTIES OF *LAVANDULA* L. GENUS PLANTS
ESSENTIAL OILS**

M.M. Gryshko National Botanical Garden, NASU, Kyiv, Ukraine.

vandae@ukr.net

The antimicrobial activity of plant oils and extracts has been recognized for many years. However, effect of essential oils on microorganisms require further researches. The influence of essential oils of *Lavandula* L. genus plants on different groups of microorganisms was shown. Screening of essential oils extracted from lavender (*Lavandula angustifolia* Mill.) and lavandin (*Lavandula hybrida* Rev.) for antimicrobial activity was done by the disk diffusion method. Different concentrations of each essential oil ranging from 0,15-5mk/ml were tested. Test cultures of soil micromycetes *Fusarium oxysporum* Schlecht., *F. solani*

(Mart.) Sacc.), *Penicillium funiculosum* Thom., *P. brevicompactum* Dierckx), *Aspergillus ochraceus* Wilhelm, *A. niger* Tiegh.) were used for experiments. Antifungal effect of essential oils of lavender and lavandin was also studied on the test cultures of pathogenic micromycetes: *Fusarium oxysporum* (Schlecht.), 50679, *Fusarium avenaceum* (Sm.) Sacc., 50706, *Fusarium culmorum* (Sm.) Sacc., 60790 та *Alternaria alternata* (Fr.) Keissler, 16858, *Alternaria tenuissima* (Fr.) Wiltshire., 16859. Fungicidal and fungistatic influence of essential oils on the growth and development of phytopathogenic micromycetes is established. *Fusarium* fungi were found to be more sensitive to the action of essential oils, and among them - *F. oxysporum*. *Lavandula hybrida* essential oil was shown to have a higher antimicrobial effect. The effect of essential oils was directly proportional to their concentration.

The antimicrobial properties of *Lavandula angustifolia* and *Lavandula hybrida* oils to phytopathogenic bacterial cultures of *Pseudomonas syringae* p.v. *syringae* 8511, *P. fluorescens* 8573, *Erwinia caratovora* subsp. *caratovora* 8982, *Xanthomonas campestris* p.v. *campestris* 8003, *Clavibacter michiganensis* subsp. *michiganensis* 102, *Agrobacterium tumefaciens* 8628 were also studied. Bactericidal and bacteriostatic effects aromatic plants oil to phytopathogenic bacteria was shown. The bactericidal effect on *Xanthomonas campestris* and, in partly, *Clavibacter michiganensis* has been established. Estimation the essential oils effect on pathogens involves the creation fungicides and herbicides.

Zaharova O. G. ¹, Tigunova O. O. ², Rahmetov J. B. ³, Shulga S. M. ⁴.

**BUTANOL ACCUMULATION BY *CLOSTRIDIUM* SP. USING
LIGNOCELLULOSE SUBSTRATES**

¹National University of Life and Environmental Sciences of Ukraine;

²Government Entity "Institute of Food Biotechnology and Genomics" of National Academy of Science of Ukraine, Kyiv;

³Kyiv National Botanical Garden named after MM Gryshko, National Academy of Sciences of Ukraine

hedg1d@gmail.com

The aim of work investigates butanol accumulation by *Clostridium* sp. using lignocellulose substrates (shredded biomass of soy, rape, wheat and switchgrass).

For the research we used: butanol producer strain *Clostridium* sp. IMB B-7570(IFBG C6H 5M) from the "Collection of microorganisms' strains and plant lines for agricultural and industrial biotechnology", Government Entity "Institute of

Food Biotechnology and Genomics of the National Academy of Science of Ukraine"; shredded green biomass of soy *Glycine max*, rape *Brassica napus*, wheat *Triticum sp* (all National Research Center "Institute of Mechanization and Electrification of Agriculture" of the National Academy of Science of Ukraine) and switchgrass *Panicum virgatum* L. (Kyiv National Botanical Garden named after M.M. Gryshko).

Cultivation of *Clostridium* sp. IMB B-7570 using lignocellulose substrates (shredded biomass of soy, rape, wheat and switchgrass) have been done and the accumulation of butanol was investigated.

Results. It was shown the largest accumulation of butanol (2,6 g/l) using shredded biomass of switchgrass as substrate and rape (2,3 g/l) and the lowest using wheat (0,2 g/l). The effect of different concentration of biomass of switchgrass and rape was conducted. It was shown the different grade of shred have effect on butanol accumulation. The largest accumulation was by shed grade of 200 mesh. In the case of preparation of the sowing material, it has been shown that the seed medium containing glycerin is optimal, and the concentration of the seed material was optimal - 10%. The largest butanol accumulation (2,0 g/l) was received using biomass rape in concentration 10 g/l.

Conclusions. The conducted studies have shown non-traditional substrates - shredded biomass of soy, rape, wheat and switchgrass have converted to biobutanol with butanol accumulation depends on substrates and seed material concentration.

Zelena L.B.¹, Ostapchuk A.M.²

DETERMINATION OF FATTY ACID COMPOSITION OF AGING YEAST COLONY

¹Zabolotny Institute of Microbiology and Virology NAS of Ukraine, Kyiv, Ukraine;

²Odessa I.I.Mechnikov National University, Odessa, Ukraine

zelenalyubov@hotmail.com

Microorganisms are able to form various structures when their cells contact to each other and combine in the complex structures. It protects them from different stresses and allows surviving in the changeable conditions.

A colony is one of the multicellular forms of microorganisms, yeasts in particular, growing on the solid surface. Cells in the colony are differentiated and specialized according to their morphological, biochemical and functional characteristics.

This study was aimed to compare whole cell fatty acids composition of yeast culturing for 21 days and assess possible differences between colony central and outer cell layers. For this purpose whole cell fatty acids composition of the cellular lipids were analyzed by the chromatography-mass spectrometry using Agilent 6890N/5973inert (Agilent Technologies, USA). *Saccharomyces cerevisiae* was cultured on YEPD agar and 24-hour, 15-day and 21-day samples were taken for analysis.

Comparison analysis of fatty acid composition showed that colonies of different ages were characterized with the highest level of 9-hexadecenoic acid (9-C16:1). The values of C12:0, C14:0 and iC15:0 were decreasing with the colony aging whereas the contents of C18:1 and iC17:0 were increasing with the maximum on 21st day of cultivation. Small amount of arachidic acid (C20:0) was detected in 15-day colony and vaccenic acid (11-C18:1) was revealed only in 21-day yeast colony.

The distinctive features of fatty acids content were observed between outer and central cell layers of *Saccharomyces cerevisiae* colony: some saturated fatty acids, such as lauric and iso-margaric, were detected only in central regions and the ratio of unsaturated to saturated fatty acids was higher in outer regions. The increasing of latter value was observed along with the colony aging. Totally, differences between outer and central cell layer were less significant than between colony of different ages.

Thus, fatty acid composition and content in yeast colony was depended on the culturing duration and colony region: fatty acids variety was narrowing with colony aging and it was more diverse in central regions.

Zelena L.B.¹, Tugay A.V.¹, Kostyuk D.², Volkova A.², Tugay T.I.^{1,2}

**STUDY OF POSSIBLE NUCLEOTIDE VARIABILITY AMONG *ASPERGILLUS*
VERSICOLOR POST-RADIATION GENERATIONS**

¹Zabolotny Institute of Microbiology and Virology, Kyiv, Ukraine;

²Open International University of Human Development "Ukraine", Kyiv, Ukraine
tatyana.tugay2@gmail.com

At present, it is urgent to study the long-term consequences of the action of chronic irradiation on the mycobiota. Particular attention needs to be paid to the study of changes in postradiation generation of micromycetes and mechanisms of trans-generation adaptation to the effects of chronic irradiation, which may lead to certain changes in the cenosis, in particular, in the rate of translocation of radionuclides in the soil. Three post radiation generations of the *Aspergillus*

versicolor were obtained in model system. The comparison was made between generations of *A. versicolor* 432 that were irradiated for the first time, its parent strain was not irradiated and did not exhibit radioadaptive properties and generations of parent strains *A. versicolor* 99 and 55 that have long existed in the Exclusion Zone and exhibited radioadaptive properties.

A. versicolor genome was analyzed by ISSR-PCR method. DNA from 3 microfungi strains was isolated and amplified with 2 primers composed of dinucleotide repeats to study possible variability of nucleotide sequences. Patterns of amplicons obtained for exposed and unexposed samples were scored and compared between various strains at the beginning and every 2 weeks of generation experiment.

Results of PCR analysis showed that the sizes of amplicons were from 200 to 1000 bp and their amount accounted up to five clear and reproducible bands. The parental strains were characterized with its particular set of PCR-products. Cultivation of *Aspergillus versicolor* strains under radioactive exposure for 2 months had different impact on microfungi genome organization and depended on the isolation source. There were no changes in amplicon patterns of strains isolated from Chernobyl region and, on the contrary, level of polymorphic bands in *A. versicolor* 432 was 25% after the first re-inoculation. Strains isolated from the contaminated soil might be characterized as adapted to chronic radiation that could explain genome invariability.

Zelena P.¹, Kharkhota M.², Yumyna Iu.¹, Faidiuk Yu.¹, Vokal S.¹, Senchylo N.¹, Skivka L.¹

**FATTY ACID FRACTIONATION ANALYSIS OF BACTERIA OF
PSEUDOMONAS AND *PANTOEAE* GENERA**

¹Taras Shevchenko National University of Kyiv, ESC "Institute of Biology and Medicine";

²Zabolotny Institute of Microbiology and Virology of NAS of Ukraine .

juliayumuna@ukr.net

Previous studies of UV-C effect on the survival of gram-negative microorganisms isolated from the bacterial epiphyte of *Oenothera sp.* growing in the 10 km exclusion zone of the Chornobyl NPP indicated the heterogeneity of epiphytic bacteria populations on the basis of sensitivity to this stressor. Along with more resistant (*Pantoea sp.* H7 and *Pseudomonas sp.* 14), there were highly UV-C sensitive variants (*Pantoea sp.* H8). Well-known is ability of bacteria to change the qualitative and quantitative composition of fatty acids in the membrane lipids under the influence of environmental stress factors. **Objective:**

To analyze the fatty acid composition of bacteria of *Pseudomonas* and *Pantoea* genera. **Results:** *Pseudomonas aeruginosa* ATCC 27853 is characterized by a predominance of oleic and palmitoleic acid fractions, which account for 35.6% and 15.9% respectively, saturation coefficient (SC) is less than 1. For *Pseudomonas* sp. 14 prevailing are palmitic (35.6%), palmitoleic (24.3%), dodecanoic (17.7%), oleic (15.6%) acids. SC: 1.81 ± 0.34 . *Pantoea* sp. H7 is characterized by a predominance of palmitic (48.1–50.8%), cyclopropaneoctanoic (12.1–14.6%), with high content of palmitoleic acid (22.7–23.2%). SC: 2.47 ± 0.1 . The dominant fraction for *Pantoea* sp. H8 is represented by palmitoleic acid (37.2–38.8%); the content of C12 and C18 fatty acids is 12.5–12.7% and 29.9–31.2% respectively. SC: 1.25 ± 0.23 . *P.agglomerans* ATCC 33248 is characterized by a predominance of palmitic (40.1–44.6%), cyclopropaneoctanoic (12.2–20.2%) acids. Also, a high content of unsaturated hexadecenoic acids fraction (19.0–27.9%), a relatively low content of saturated medium-chain fatty acids (C12:0 + C14:0) fraction, of 12.9–14.1%, are observed. SC: 2.85 ± 0.62 . **Conclusions:** A clear correlation between UV resistance and saturation coefficient is shown: the most resistant strains are characterized by the predominance of saturated fatty acids, while sensitive — by unsaturated. Decrease of unsaturated fatty acids content should reduce the number of sensitive sites (unsaturated bonds) that are highly oxidizable by active forms of oxygen that emerge in cells due to radiation or ultraviolet influence.

Zelena P.P¹, Shepelevych V.V¹., Stepura L.G¹., Yumina Yu. M¹., Dzyublyuk N.A²

**SPECIAL FEATURES IN SPORE-FORMING BACTERIA, ISOLATED FROM
THE PERIPHERY OF AVIARIES *DELPHINUS DELPHIS*.**

¹Kyiv National University of Taras Shevchenko, Kyiv, Ukraine.

larisastepura@ukr.net

Spore-forming bacteria of the genus *Bacillus* is a promising group on the study of their properties in order to introduce these bacteria into further development in various fields: food industry, genetic engineering, biotechnology. The aim of the work is to discover biological properties of aerobic spore-forming bacteria isolated from periphery of aviaries. In this work microbiological, biochemical, microscopic methods of investigation were used.

It is established that the studied microorganisms are mobile, gram-positive, with cell diameter less than 1 μm , formed ellipsoid or cylindrical (strains of bacteria 2.1) central / paracentral spores, did not form parasporic crystals and

capsules, did not accumulate poly- β -hydroxybutyric acid, catalase-positive and oxidazonegative. All investigated cultures had an optional anaerobic type of metabolism, split glucose with the formation of acetoin, were able to use sodium citrate and many other organic compounds as the sole source of carbon. Bacteria of strains 1.2.1, 1.2.r, 2.2.1 reduced nitrates to nitrites, strain 2.1 - to nitrite and nitrite. The isolated microorganisms did not split the sulfur-containing amino acids and tryptophan, but utilized the protein compounds with ammonia release, the urease was detected in strains 1.2.1 and 2.2.1. Bacteria of strains 1.2.1, 1.2.r, 2.1, and 2.2.1 are positive for arginine dihydrolase and negative for ornithine decarboxylase; bacteria of strains 1.2.1 and 2.2.1 also synthesized lysedecarboxylase, strains 2.2.0 cells are not able to decarboxylize arginine, lysine and ornithine. It is established that the bacteria studied are capable of hydrolyzing casein, gelatin, esculin and carboxymethyl cellulase. Strains of 1.2.1, 1.2.r, 2.1 and 2.2.1 produced the hydrolysis of starch and olive oil.. Isolated microorganisms grown at a concentration of NaCl up to 7.5%; pH 5.7 - 8.0; temperature 28⁰+C - 50⁰+C. Thus, as a result of the study on phenotypic characteristics of the culture of bacteria 1.2.1, 1.2.r, 2.1 and 2.2.1, the properties are characteristic of *Bacillus licheniformis*.

Zhhanynch I.P., Sharga B.M.

**STUDY OF FUNGICIDES SPRAYS CONCENTRATION EFFECT ONTO
WINTER WHEAT FUNGAL INFECTIONS IN TRANSCARPATHIA**

Uzhhorod National University

bmsarga@yahoo.co.uk

Same treatments of crop near Uzhgorod were applied to control or experimental plots, however, for latter we decreased the doses of fungicides, mentioned below for control, twice.

Seeds treatment: fungicide Lamardor Pro 0.6 L/t + insecticide Gaucho Plus 0.6 L/t.

Spring spray 1: fungicide Bumper super (propiconazole 90, prochloraz 400 g/l), 1L/ha, herbicide Grodil Max (bentazone 352.4 + Acyfluorfen 161.7 g/L), 0.11 L/ha. The carbamide (10 kg/ha) was used in both treatment variants.

Treatment 2: growth regulator Ceron (Ethephon 480 g/l), 1 L/ha, the insecticide Decis Profi (Deltamethrin 250 g/kg), 0.04 Kg/ha and fertilizer «Опакул мультикомплекс» (microelements), 1 L/ha.

Treatment 3: insecticide Pirinex super CE (chlorpyrifos 400 and bifenthrin 20 g/L), fungicides Thesis ALFA Smart Agro (tebuconazole, 500 g/l), 0.5 kg/ha and

Grinfort (Cyproconazole + propiconazole 250g/L), 0.5 L/ha. Then, carbamide (5 kg/ha) was casted to all plots.

Detected diseases: Fusarium foot rot/head blight; *Puccinia* spp. rusts; powdery mildew (*Erysiphe graminis* f. sp. tritici); black point disease (*Alternaria* spp., *Cochliobolus sativus*, *Cladosporium* spp.) were most prevalent and more severe in plots treated by less concentrated fungicides.

Fusariosis appeared at mid-summer in small numbers on well treated plots and in about 5 times more large quantities onto fields with half-dose sprays.

Yellow rust, *Puccinia striiformis* f. sp. tritici showed weak symptoms and lowest incidence among the rusts on poorly treated plots and only few diseased plants on properly treated fields.

P. recondita and *P. graminis* f. sp. tritici infections were of moderate severity/spread on bad treated plots and with little symptoms/incidences on well sprayed fields.

The powdery mildew severity was scored 1 on well treated and 3 on less treated plots. *Alternaria* spp., *Cochliobolus sativus*, *Cladosporium* spp. were detected in black dots at harvest. Black point disease was found in < 2% of grains on well sprayed plots and in > 9% of grains from less concentrated sprays plots. Smut (*Ustilago tritici* Jens) was present in about 5% of plants only on fields treated by halved doses of fungicides.

Zhornyak O.I., Zhornyak P.V.

ANTISEPTIC MEDICINAL PREPARATION ANTIMICROBAL ACTIVITY RESEARCH

National Pirogov Memorial Medical University, Vinnytsya

zhornjak.ei@gmail.com

Modern problems of fight with microorganisms resistant cultures demand constant search of new antimicrobial methods and their influence study on resistant bacteria forms. Antiseptic methods are widely used in medical practice for mouth and throat mucous cavity inflammation processes treatment. In list of those methods absolute leadership belongs to pill forms for resorption. Among basic active substances pill mixture amilmetakrezol, decamethoxin, benzolkoniy chlorid and other antiseptics are used. Presence of quite variegated methods arsenal with antiseptics of different chemical groups, absence of comparative research and their action results information, mouth cavity microflora resistance formation, collateral influence on the human organism make difficult rational choice of the method in every specific case.

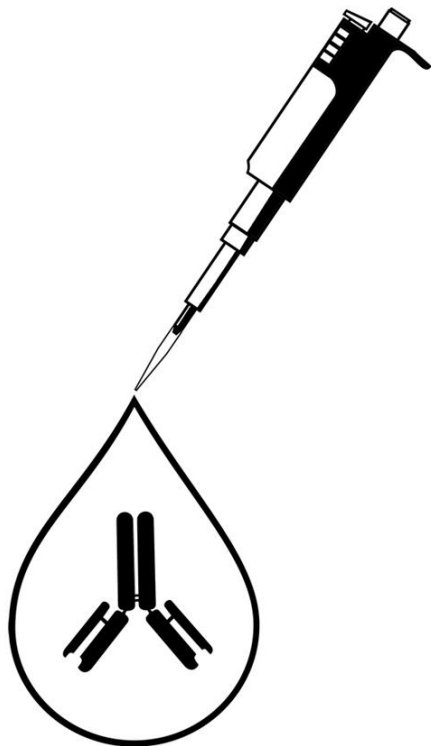
For the decision of a set purpose, a study of antimicrobial pill Ephisol (dekvalin chlorid), Adzhysept (amilmetakrezol), Septefril (decamethoxin) properties was carried out in clinical and museum staphylococcus cultures.

Antimicrobial action preparation research was done by the generally accepted method of serial double sequential cultivation. Pills were previously dissolved by steril distilled water to 5 ml volume. Results of the research testify that method of serial sequential cultivation in liquid nourishing environment allows to establish minimal active antiseptics concentration and gives a possibility to judge of microorganisms sensibility level towards researched preparation, taking into account effective medicine concentration creation.

It was established that preparations: septefril (decamethoxin) showed baneful influence on staphylococcus cultures. Adzhysept, Ephisol showed lower effectiveness on staphylococcus.

So, pill antiseptic preparations have bactericidal action to staphylococcus cultures. During action comparison, the most effective appeared to be antiseptic preparations Septefril.

SECTION



IMMUNOLOGY

**Akimova V.M., Lapovets N.E., Lutsiv N.Z., Martianova O.I., Lebed G.B.,
Lapovets L.E.**

**SERUM LEVEL OF TNF α AND SOLUBLE TNFRP55 IN ABDOMINAL
TUBERCULOSIS**

Danylo Halytsky Lviv National Medical University, Lviv, Ukraine

viorikakimova@gmail.com

Abdominal tuberculosis (AT) can have a varied clinical presentation, frequently mimicking "acute abdomen", in particular, nonspecific acute mesenteric lymphadenitis (AML). So, it is important to find laboratory criteria for differential diagnosis of AML and AT. In pathogenesis of the AML and AT is directly involved immune system. TNF-mediated signaling through TNF receptors contributes to protective immunity against Mycobacterium tuberculosis. TNF receptors can be in soluble form after membrane shedding. The effect of soluble TNFRp55 is controversial. Evidence proposing an antagonistic and agonistic effects.

Aim. To determine serum concentration of TNF α and soluble TNFRp55 in patients with AML and AT.

Methods. This study investigated the inflammatory response pattern of TNF α and its soluble receptor in patients with AML (n=27), AT (n=22) and 25 healthy people. Peripheral blood serum were analyzed with ELISA for concentration of TNF α and soluble form of TNF receptor (sTNFp55) preoperatively.

Results. The serum level of TNF α was significantly higher in the AT (17.6 ± 1.1 pg/ml) than in control group (4.9 ± 0.1 pg/ml) ($p < 0.05$). In patients with AT TNF α was 3 times higher than with AML (5.6 ± 0.2 pg/ml).

The serum level of sTNFp55 was higher in AML group (3.0 ± 0.2 ng/ml) and AT (3.5 ± 0.1 ng/ml) than in control (2.2 ± 0.2 ng/ml) ($p < 0.001$). The sTNFp55/TNF α ratio was higher in AML group (530.6 ± 2.6) and in AT (201.2 ± 10.2) was lower than in control (432.6 ± 20.1).

Conclusion. The increase of sTNF-R1 serum level in AML may be compensatory to bind serum TNF α to prevent destructive action of it. And this is possible pathophysiological role of TNF and soluble receptors in local inflammatory process. In abdominal tuberculosis TNF α secretion is prevailing over the sTNF-R1 shedding, and this may play role in pathogenesis of tuberculosis. Measurement of serum TNF α and sTNF-R1 level can be useful in differential diagnosis of AML and AT.

**Alexandrov A.V., Konopelnuk V.I., Kompanets I.V., Holoborodko Ye.Ye,
Svyatetska V.M., Molozhavaya O.S., Ostapchenko L.I.**

**THE EFFECT MELANIN ON PROINFLAMMATORY STATUS OF PERITONEAL
MACROPHAGES OF RATS WITH PROGESTERONE-INDUCED OBESITY**

Taras Shevchenko National University of Kyiv, Kyiv, Ukraine

ir_kom@ukr.net

The systemic inflammation following the local low-grade inflammation in adipose tissue is one of general outcomes of obesity. Inflammation also triggers the oxidative stress: pro-inflammatory cytokines promote increased production of reactive oxygen (ROS) and nitrogen (NO) species by macrophages and monocytes. Little data are available regarding the role of peritoneal macrophages in inflammation in obesity. The mechanism of obesity induced by progesterone used for contraception and hormone replacement therapy remains poorly understood.

Therefore, inflammatory reactions could be the treatment target in obesity. In this regard pigment melanin produced by Antarctic black yeast may have beneficial effect on inflammation in obesity. Its antioxidant and anti-inflammatory properties have been demonstrated.

The present study was designed to explore the effects of melanin (Mel) from Antarctic yeast *Nadsoniella nigra* on NO and ROS production by peritoneal macrophage of rats with progesterone-induced (Prog) obesity.

A 45% increase of NO level was observed in peritoneal macrophages of Prog-treated rats in comparison to the control group. Mel administration to Prog-treated rats led to more than twofold decrease of NO level as compared with the group treated with Prog only, it was even lower than control. Notably, Mel exerts more powerful effect on NO production by macrophages of obese animals as opposed to healthy ones.

Unexpectedly, no significant elevation in ROS level in peritoneal macrophages has been observed in healthy and Prog-treated rats indicating that oxidative stress are not implicated in response of these cells to inflammation in obesity. ROS level in Prog+Mel group decreased 9% in comparison with animals, treated with Prog only. Notably, the ROS level in Prog+Mel group was 9% lower than the control, like that in control group receiving Mel.

We suppose that peritoneal macrophages contribute to the inflammatory response in Prog-induced obesity more by the production of NO than by ROS generation due to oxidative stress. Mel prevents macrophage activation in

response to low-grade inflammation in obesity and may have beneficial effect on obesity prophylaxis.

Aminov R.F., Frolov O.K.

FUNCTIONAL STATE OF T-LYMPHOCYTE RATS IN VITRO UNDER THE
INFLUENCE OF BIOLOGICALLY ACTIVE SUBSTANCES OF SALIVES OF
HIRUDO VERBANA IN VIVO

Zaporizhzhya National University, Zaporizhzhya, Ukraine

91_amin_91@ukr.net

Hirudotherapy is widely used in medicine and veterinary medicine, which has many therapeutic effects, due to the introduction of more than 100 substances into the host organism, in different directions of action, which are dosed in sequence. However, this therapeutic method remains empirical, due to insufficient study of mechanisms of action, especially immunotropic. Therefore, it was important to investigate the potential proliferative activity of lymphocytes and their sensitization to BAS in the culture of blood lymphocytes in the reaction of blast transformation of lymphocytes (RBTL), which would respond to the immunological effects of hirudotherapy and dosage it according to the principles of protocol medicine. Sexually mature nonlinear females of rats, two weeks prior to pairing and two weeks afterwards were examined, one hungry medical leeches *Hirudo verbana* were consecutively put each week for a month. The females were studied after feeding the offspring, as well as the offspring for the 45th and 60th day. After decapitation of rats under anesthesia, blood was taken, it was diluted with heparin (1:10), nutrient medium (1: 5). The culture mixture was injected by mitogen - concanavalin A (Con A). Salt extract from bodies *H. verbana* was used as antigens, cultured for 24 hours at a temperature of +37. After it has cultured for 24 hours, the samples were centrifuged, the culture supernatant was harvested, and preparations were prepared from the cell, fixed and stained. The level of RBTL was evaluated morphologically, taking into account 300-400 lymphocytes. As a result of the experiment on the background of the hirudological influence on the stimulation of whole blood by the antigens of the medical leech, the percentage of blast cells significantly increases in both sexually mature female rats and in their offspring. The RBTL indicators for *Hirudo verbana* are increased and approached to indicators, as on plant lectin ConA, that indicating their polyclonal activation. The result is an immunostimulating effect on the acquired part of the immune system of animals.

Bakhmachuk A.O.^{1,2}, Gorbatiuk O.B.^{1,3}, Rachkov A.E.¹, Soldatkin A.P.^{1,2}
DEVELOPMENT OF SPR IMMUNOSENSOR USING INTERMEDIATE LAYER
BASED ON THE RECOMBINANT STAPHYLOCOCCAL PROTEIN A
IMMOBILIZED THROUGH HIS-TAG

¹Institute of Molecular Biology and Genetics, NAS of Ukraine, Kyiv, Ukraine;

²Institute of High Technologies, Taras Shevchenko National University of Kyiv, Kyiv, Ukraine;

³State Institute of Genetic and Regenerative Medicine NAMS Ukraine, Kyiv, Ukraine
a.bakhmachuk@gmail.com

In comparison to the existing conventional analytical approaches, biosensors have several advantages: they provide easy, fast, accurate, highly sensitive, specific, and cheap measurements. The antigen-binding activity of immobilized antibodies is usually much lower in comparison with the same soluble antibody activity in due to the random orientation of antibodies on the sensor surface and steric hindrance caused by the influence of the surface of a solid substrate. To prevent this, an intermediate layer of immunoglobulin-binding Staphylococcal surface protein A (SPA) that selectively binds Fc fragment of antibody leaving Fab fragment available for antigen detection, can be used. However, SPA itself also meets the same problem of random immobilization. To facilitate more effective immobilization of the protein on a gold sensor surface, the recombinant SPA, containing His-tag, was used. Thus, the aim of this work was to investigate the possibility of using the recombinant SPA immobilization on a sensor surface through His-tag in order to form intermediate layer of immunoglobulin-binding proteins for further development of immunosensor.

The study of the process of immobilization and intermolecular interactions between immune components was performed by using the measuring flow cell of the surface plasmon resonance (SPR) spectrometer "Plasmon-4m".

A successful immobilization of recombinant SPA on the gold sensor surface through His-tag using analogue of NTA molecule (BCL-12) was performed. The dependence of the sensor response at the interactions between immobilized recombinant SPA and rabbit antibodies against lactoferrin (anti-LF) on the anti-LF concentration as well as on lactoferrin (LF) concentration at the interactions between anti-LF and LF were shown. Some biosensor characteristics, like storage stability, reproducibility and sensitivity, will be discussed.

The obtained results show that the formation of intermediate layer of recombinant SPA through its His-tag is a promising way for development of immunosensor.

Baranova G.V., Zatovska T.V., Golovan A.V., Zagorodnya S.D.

**USE OF SPR ANALYSIS FOR DETECTION OF SPECIFIC ANTIBODIES
AGAINST HERPESVIRUSES IN HUMAN BLOOD SERUM AND RESEARCH OF
THE BINDING OF DNA TO ANTIVIRAL DRUGS.**

Zabolotny Institute of Microbiology and Virology of NASU, Kyiv, Ukraine.

tzatovska@ukr.net

The development of biosensor technologies based on the phenomenon of surface plasmon resonance (SPR) is one of the priority directions in bioanalytics. The application of biosensors allows you to analyze the binding of biomolecules at very low concentrations in real biological probes, without the use of any labels. Optical biosensors are used in clinical analysis to detect hormones, specific markers of cancer and cardiovascular diseases, specific antibodies to viruses and bacteria in human blood. Previously, biochips for the rapid detection of specific antibodies to HSV-1 and EBV were developed in our Department. The prepared biochips were suitable for SPR analysis for 6 months. The presence of specific antibodies in human serum was determined by the resonance response, the time of analysis was about 20 min. The biochip could be restored with a dissociating solution and used for the analysis of various sera at least three times. Laboratory tests of developed immunosensory test systems for the detection of specific IgG against viruses with the use of serum blood of patients with various diagnoses, conducted at the Gromashevsky Institute of Epidemiology and Infectious Diseases of NAMS of Ukraine confirmed their effectiveness and specificity

Another direction of SPR analysis is the development of biosensors for the study of nucleic acid interactions. The use of DNA sensors in pharmacology promotes the search for new effective antiviral and antitumor drugs. The data on the modification of the biochip surface for sorption of viral DNA and a series of correlation relationships between the mass of sorbed DNA and the response of the sensory system had been obtained. The ability of sorbed DNA to bind to potential DNA by intercalators has been established as the basis for screening of compounds with probable antiviral properties. Thus, SPR analysis can be used to study the binding of DNA with drugs that are tested on antiviral activity in cell cultures.

Bilokur D. O.

**IMMUNE STATUS OF INDIVIDUALS IN THE TERRITORIES OF INTENSIFIED
RADIOECOLOGICAL CONTROL OF THE SUMY REGION**

A.S. Makarenko Sumy State Pedagogical University of Ukraine, Sumy, Ukraine
darina.bilokur@gmail.com

All of the test parameters were within the clinical norms. Individuals from Sumy intensified radioecological territories have been observed to decrease the absolute amount of leukocytes and neutrophils comparing with the control group. There is an increase in absolute and relative values of indicators of eosinophils, basophils and monocytes (by 2,6, 7,5 and 1,73 times correspondingly).

The relative number of lymphocytes is within the clinical norm and is close to the medical indicators of the control group. The absolute number of lymphocytes tends to decrease.

It was pointed out the decrease of absolute and relative rates of T-lymphocytes with phenotypes: CD3 (in 1,5 times), CD4 (in 1,34 times) and NK CD16 - in 2,3 times. The number of T-lymphocytes with the CD8 phenotype was within the limits of clinical norms and did not differentiate from the number of the control group.

There is a 1.5-fold decrease in the immune-regulatory index (IRI).

It was established that the relative and absolute number of B-lymphocytes along with the CD22 phenotype was higher than the control one. There is a decrease in the concentration of Ig G, an increase in the concentration of Ig A and Ig M relative to the control values. At the same time, the Ig G, Ig A indices did not go beyond the clinical norm. Instead of the concentration of Ig M was higher by 3.75 times accordingly to the control group indices and by 1.26 times, relative to the upper limit of the clinical norm.

Thus, the obtained results indicate the functional load of the cellular and humoral parts of systemic immune system among the population of radiation-polluted territories of the Sumy region. At the same time, the formation of compensatory mechanisms in response to low-intensity prolonged radiation irradiation is observed.

Bolshakova K.M.¹, Dons'koi, B.V.², Chernyshov, V.P.²

**DIVERSE SIGNIFICANCE OF CD8 EXPRESSION ON NK CELLS FOR THEIR
CYTOTOXICITY AGAINST HLA^{POS} AND HLA^{NEG} TUMOR CELL LINES**

¹Taras Shevchenko National University of Kyiv, Kyiv, Ukraine;

² Institute of Pediatrics, Obstetrics and Gynecology, National Academy of Medical Sciences of Ukraine, Kyiv, Ukraine

bolshakova.kateryna@gmail.com

Natural killer (NK) cell cytotoxicity has an important role for embryo implantation and placentation. Cytotoxicity of NK cells (NKc) is measured against K-562 cell lines as "gold-standart" target cells. In contrast, tumor cell lines Jurkat and Molt-4 expresses amounts of Human Leukocyte Antigens (HLA) Class I and are more resistant to NKc. Effectiveness of resistance highly depends on interaction between NK-receptors and HLA on target cells. This topic requires a better understanding.

Methods: Donors blood samples was analyzed for NKc and activity (stimulating CD69 expression) against 3 tumor cell lines: K-562, Jurkat and Molt-4, by flow cytometry. Also NK phenotypes (CD8, CD158, HLA-DR) were analyzed using monoclonal antibodies (BD Biosciences).

Results: Levels of NKc against Jurkat and Molt-4 (18% and 16%) was lower than to K-562 (24%). But we've found a significant correlation of NKc between all 3 lines. NKc against K-562 has an extremely significant correlation with cytotoxicity against Jurkat and Molt ((r) = 0.7070, (r) = 0.8275). We've found that NKc against K-562 and Molt-4 correlate with NK% (r = 0,465133, r = 0,624574). It was shown that NK CD8 phenotype influenced NKc against different cell lines. In case of NKc against K-562, CD8^{pos}NK% correlate with NKc (r = 0,532049). In contrast only CD8^{neg}NK% correlate with NKc against Molt-4 (r = 0,704875). Moreover only DR^{neg}NK numbers (but not DR^{pos}) correlate with NKc in all 3 lines. NKc against all 3 lines hadn't depended from CD158 NK expression.

Conclusion: We've shown that NK phenotype determine the effectiveness of response against specific cell line. CD8^{pos}NK cells are more effective in killing HLA^{neg} K562. But in same time they don't take significant part in killing HLA^{pos} Molt-4. It supports the idea that possible role of CD8 on NK is the recognition of HLA.

Bychkova N.G., Bychkov O.A., Tarasyuk A.P.

**ADHESION DISORDERS IN DEVELOPMENT OF COMBINED ARTERIAL
HYPERTENSION AND GOUT**

National Medical University named after O.O. Bogomolets, Kyiv, Ukraine

oleg_bichkov@yahoo.com

The purpose of the work. Studying features of immune status in patients with combined hypertension and gout.

Materials and methods. The study involved examination of 137 male patients with stage II hypertension, average age 56.9 ± 3.4 . All patients underwent echocardiography with estimation of the left ventricular mass index to verify hypertension stage, blood chemistry test with estimation of uric acid level, as well as lipid profile and immune status. The control group included 35 healthy subjects randomized by age and gender.

As a **result** of the research we found that the main group of patients demonstrated positively higher content of activated lymphocyte subpopulations carrying various cell adhesion molecules and receptors thereof. For instance, content of CD54+ lymphocytes expressing cell adhesion molecule ICAM-1 exceeded the comparison group value by 27.15% ($p < 0.05$), CD11b - by 38.29% ($p < 0.05$), CD62L - by 13.76% ($p < 0.05$). It is CD11b receptor that provides for adhesion of macrophages and neutrophils to the endothelial wall and acts as a ligand for ICAM-1, CD62L (L-selectin) provides for adhesion and adherence of lymphocytes to the endothelial wall.

We have also found high serum concentration of soluble vascular cell adhesion molecule sVCAM, the level of which in the main group exceeded the similar value in the comparison group by 42.75% ($p < 0.05$), sICAM-1 - by 18.61% ($p < 0.05$). Patients with combined abnormalities, as well as patients with isolated hypertension demonstrated positively higher content of CD30+lymphocytes by 51.7% ($p < 0.05$) and by 49.4% ($p < 0.05$) respectively.

Conclusion. Positively higher percentage of activated T-cells was found in patients with combined hypertension and gout, both with early and late activation marker, as well as those expressing FAS receptor, and ready to enter into apoptosis. We have identified abnormalities in adhesion and cooperation of immune competent cells, which indicates type 2 T-helper immune response prevailing in atherosclerosis-associated diseases and development of autoimmune disorders associated with endothelium dysfunction and development of atherosclerotic inflammation.

Bychkova S.A.

INTERLEUKIN-17, INTERFERON- γ AND TRANSFORMING GROWTH FACTOR- β DISTURBANCES IN PATIENTS WITH DIFFERENT PHENOTYPES OF CHRONIC OBSTRUCTIVE PULMONARY DISEASE

Ukrainian military medical academy

svetlana_bichkova@yahoo.com

Purpose is - to study of immunologic features of the disease course of different COPD phenotypes – with chronic bronchitis and emphysema, and role of interleukin-17 (IL-17), interferon- γ (IFN- γ) and transforming growth factor- β (TGF- β) in the pathogenesis of chronic obstructive pulmonary disease (COPD). 87 male patients were involved in the examination, informed consents of all of them were obtained. Average age of patients was $53,9 \pm 4,5$ years. Control group consisted of 35 persons, randomized by age and sex without any signs of COPD. Levels of pro- and anti-inflammatory cytokines, IL-17A, interferon- γ (IFN- γ), transforming growth factor- β (TGF- β) and IL-21 by "Genzyme diagnostics". For the preparation of supernatants with IL-17, TGF- β and IFN- γ cells of peripheral blood (heparinized blood diluted 1:4 was used) were incubated in the presence of a mitogen (LPS-induced synthesis) and in the cultivation medium (RPMI-1640 without a mitogen – spontaneous synthesis) for 24-48 hours in the atmosphere with 5% CO₂. Patients with chronic bronchitis phenotype were observed to have signs of metabolic syndrome, lipid metabolism disorders and insulin resistance, unlike patients with emphysema phenotype. Patients of both clinical phenotypes have an equal severity of airway obstruction, exacerbation frequency and systemic arterial hypertension as a concomitant disease.

Patients with COPD with phenotype of chronic bronchitis and frequent exacerbations have Th17 response predomination with significant elevation of IL-17, TGF- β , IL-6 concentrations in serum and high concentration of soluble adhesion molecules along with high expression of receptors to adhesion molecules on activated lymphocytes of peripheral blood. Patients with COPD emphysema phenotype with frequent exacerbations in immune system have Th1 response predomination with significantly higher concentrations of IFN- γ and lower levels of IL-4 and IL-10, along with high level of activated lymphocytes with CD25+ phenotype and low with CD30+ phenotype. In both groups of patients with chronic obstructive pulmonary disease a high serum level of proinflammatory cytokines (TNF- α , IL-1 β and IL-8) was observed.

Demchina O.V.¹, Dmytrukha N.M.²

**RESEARCH ON THE DISINFECTANT'S INFLUENCE ON THE LIVING
CONDITIONS OF PERITONEAL MACROFAGS OF RATS WISTAR**

¹Kiev National Taras Shevchenko University, Kiev, Ukraine;

²SI "Kundiev Institute of Occupational Health of the NAMS of Ukraine", Kiev, Ukraine

olyadem4ik@gmail.com

Modern disinfectants have a wide range of uses. They are composed of various active substances that possess antimicrobial and antifungal properties. The main requirement for disinfectants is their effectiveness in relation to pathogenic microflora, safety for human health and the environment.

When disinfectants enter the human body, they can affect the normal microflora and immunocompetent cells that are involved in the formation of immunological reactions. In view of the above, the issue of the disinfectant's influence on the immune reactivity of the body is very relevant.

The purpose of the study was to evaluate the influence of disinfectants on macrophages (cells of the first line of the organism's protection from exogenous factors). The cytotoxic action of disinfectants was investigated: Bioblisk, Blisk, Blisk Plus, Max Activ (manufacturer HIGIENIQUE, Poland). Macrophages were isolated from the peritoneal exudate of the intact rats Wistar, and at a concentration of 2×10^6 cells/ml in RPMI 1640 medium were transferred to the plate. Disinfectants at concentrations of 0.1%, 0.05% and 0.025% were added to cells and incubated for 24 hours. Cell viability was tested with trypan blue dye, MTT-test and neutral red dye according to guidelines. The optical density of the samples was measured on a Sunrise Tekan device (Austria) at a wavelength of 540 nm.

The study of cytotoxic action of disinfectants showed that the largest cytotoxic activity in relation to macrophages at a concentration of 0.1% was demonstrated by Bioblisk (52% living cells), the smallest was demonstrated by Max Activ (100% living cells) according to the MTT-test and test with neutral red dye. The results of the test using neutral red dye showed that the disinfectants Blisk, Blisk Plus at concentrations of 0.05% and 0.025% demonstrated moderate cytotoxic activity (living cells were 72% and 70%).

Consequently, solutions of disinfectants in concentration of 0.1% exhibit moderate cytotoxic effects on the macrophages of rats which was shown as a violation of cell membrane integrity and mitochondrial function.

Dovgiy R.S.¹, Nikolsky I.S.², Skivka L.M.³, Butenko G.M.¹

**AGE-RELATED CHANGES IN ARGININE METABOLISM OF MURINE
MONOCYTE-DERIVED AND TISSUE-RESIDENT MACROPHAGES.**

¹D.F. Chebotarev State Institute of Gerontology NAMS of Ukraine, Kyiv, Ukraine;

²State Institute of Genetic and Regenerative Medicine NAMS of Ukraine, Kyiv, Ukraine;

³Taras Shevchenko National University of Kyiv, Kyiv, Ukraine

roman_dovhyi@univ.kiev.ua

The metabolic polarization of phagocytes largely determines their functional status. It is known that arginine metabolism of macrophages (M ϕ) changes upon aging. However, very few studies on this subject exist. Research conducted over the past decade significantly expanded the knowledge about M ϕ . It was shown that tissue-resident M ϕ differ from monocyte-derived M ϕ by their origin. Thus, the aim of this study was to compare age-related changes in the metabolic status of murine monocyte-derived and tissue-resident macrophages.

Young (2-5 mo old) and aged (18-24 mo old) C57Bl/6 mice were used in studies. Monocyte-derived M ϕ were differentiated from bone marrow monocytes in M-CSF-containing medium. We studied alveolar and peritoneal M ϕ as representatives of tissue-resident M ϕ . Alveolar M ϕ were obtained using bronchoalveolar lavage procedure. Resident peritoneal M ϕ were isolated by flushing peritoneal cavity with cold DPBS. M ϕ were enriched by adhesion to culture plastic. Griess colorimetric reaction was performed to analyze nitric oxide (NO) production. Arginase activity was determined by measuring the production of urea using spectrophotometry.

Arginase activity of monocyte-derived M ϕ obtained from old mice was significantly lower as compared to young animals, while NO production was nearly the same in different age groups of mice. Conversely, arginase activity of both analyzed types of tissue-resident M ϕ was significantly higher in old animals. There was also decrease in NO production in alveolar M ϕ from old mice compared to young, while peritoneal M ϕ from mice of different age groups didn't differ in this parameter.

Observed decrease in arginase activity indicates proinflammatory metabolic skew in monocyte-derived M ϕ from aged mice. It may serve as one of the explanations of higher incidence of systemic inflammatory diseases upon aging. Conversely, tissue-resident M ϕ from old animals had higher arginase activity suggesting their anti-inflammatory metabolic profile. Inappropriate anti-inflammatory metabolic skew of these cells may impair their patrolling function

and cause the development of malignant tumors, atopic dermatitis and fibrosis observed during aging.

Frolov A.K., Aminov R.F., Fedotov Ye. R., Litvinenko R.A.

METHODOLOGY FOR MORPHOGENETIC FUNCTION OF IMMUNITY

Zaporizhzhya National University, Zaporizhzhya, Ukraine

a_frolov@ukr.net

It has been experimentally shown that all clones of T- and B-lymphocytes are moderate autoclones (Poletaev et al., 2012), and the relationship of the immune system (IS) to "non-self" depends on the degree of its danger, according to the theory of Matzinger (2002). The discovery of patterns and receptors to them on the cells of the IS promoted an understanding of the functions integration of innate and adaptive immunity, and the heterophilicity of patterns and the cross-reactivity of receptors on them, led to the relativity of concepts "self-non-self". In this connection, the phylogenetic significance of the homeostatic function of the IS consists of morphogenetic function that is directed on the control and regulation of metabolism, proliferation and differentiation of cells of all body tissues, according to its genotype, and cytotoxic function that eliminates deviations of histogenesis (oncogenesis) and various pathogens.

Currently, the methodology for cytotoxic function of the IS is well supplied, whereas the methodology for morphogenetic function is just beginning to be developed. The main reason is that molecular genetic mechanisms have not been studied. In this connection, immunogenesis approach to determining the functional state of the IS is promising, that based on the definition of the pool of activated lymphocytes among those circulating in the internal environment of the body. The definition of activated lymphocytes is based on testing the residual signs of previous immunogenesis in the organs of the IS on circulating lymphocytes. We have identified the following informative signs of activated lymphocytes: 1) cytogenetic - according to the location of the nucleolus-forming chromosomes; 2) cytomorphometric - by the ratio of small, medium and large lymphocytes; 3) rosette – on the density of differentiating CD-structures; 4) luminescent – by the ratio of DNA and RNA in cells. These signs reflect certain stages of the preceding immunogenesis: activation, proliferation, differentiation, peculiarity of migration to specific organs. The proposed approach suggested by us allows to personalize the assessment of the IS state at the moment of examination.

**Gakhramanova M.¹, Molozhava O.², Svyatetska V.², Ostapchuk A.³,
Skivka L.²**

**PHYTOCHEMICAL ANALYSIS AND IMMUNOMODULATORY EFFECT OF
POLYHERBAL PREPARATION WITH HEPATOPROTECTIVE, CHOLERETIC
AND ANTI-INFLAMMATORY ACTIVITY *IN VITRO***

¹"NARGIZ" Medical Clinic, Baku, Azerbaijan;

²Taras Shevchenko national university of Kyiv, Ukraine;

³Odessa Mechnykov National University, Odesa, Ukraine

nergizmedical@mail.ru

The main constituent of polyherbal preparation (PP) with hepatoprotective, choleretic and anti-inflammatory activity is *Portulaca oleracea* L. (purslane). Purslane is used extensively in dietary intake around the Mediterranean and Asian countries. It is an important medicine plant in a folk medicine in many countries. *P. oleracea* exerts a wide spectrum of pharmacological effects: neuroprotective, antimicrobial, antidiabetic, antioxidant, anti-inflammatory, anticancer etc. Traditionally, *P. oleraceae* is used as a monoherbal preparation. Data concerning the use of purslane in combination with other plants are sparse. Medicinal plants used in the proposed PP are known to possess anti-inflammatory activity: *Calendula*, *Polygonum aviculare*, *Hypericum*, *Taraxacum officinale*, *Helichrysum*, *Mentha piperita*, *Matricaria chamomilla* L. This study was aimed to perform phytochemical analysis of PP and to investigate the effect of its aqueous extract on rat peritoneal macrophage (PM) viability and oxidative metabolism *in vitro*. All plant species were collected from different regions of Azerbaijan. Crude aqueous extract (CAE) was prepared by adding 100 ml of boiling water to 5 g of plant mixture followed by brewing for 30 min. Prepared decoct was then filtered and dried under vacuum in a rotary evaporator. The residue was freeze-dried in liophilizer and stored at -20°C until used. Fatty acid analysis was performed by Gas Chromatography–Mass Spectrometry. PM viability was determined in MTT-test, oxidative metabolism – in NBT-test. Qualitative phytochemical studies revealed tannins, saponins, flavonoids, water-soluble and water insoluble phenolic compounds, cardiac glycosides and cumarins. Total phenolics was $0,165 \pm 0,036$ mg gallic acid equivalent/g dried weight, flavonoids - 16 mg rutin equivalents /g dried weight, and tannins - 0,334 mg catechin equivalents /g dried weight. Most predominant fatty acids were octadecadienoic (11,46 mg/g), octadecatrienoic (10,61 mg/g) and hexadecanoic (7,04 mg/g). No cytotoxic effect in PM was detected. Treatment PM with CAE resulted in increase generation of reactive oxygen species – important signaling molecules in the resolution of inflammation.

Galkin O.Yu., Lytsenko T.M.

**BIOTECHNOLOGY OF PREPARATIONS OF HUMAN RECOMBINANT
INTERLEUKIN-7**

¹Igor Sikorsky Kyiv Polytechnic Institute, Kyiv, Ukraine;

²UA Pro-Pharma LLC, Kyiv, Ukraine

alexftb@gmail.com

The purpose of the work is the scientific substantiation of biotechnology of preparations of human recombinant interleukin-7 (rIL-7), as well as its standardization (technological, analytical).

We have developed and substantiated the optimal parameters of biotechnology for the preparation and purification of rIL-7. For the first time, it has been shown that rIL-7 effectively inhibits the reproduction of a surrogate hepatitis C virus (HCV) in vitro (CC₅₀ = 3 µg/ml, ED₅₀ = 4.7 ng/ml, IS = 640). The highest proliferation of intact T cells is determined at doses of rIL-7 0.3 and 0.025 µg/ml. rIL-7 had a different effect on infected HCV culture: in the first 3 days the number of cells decreased or did not change, and in 2-3 weeks - increased almost 2 times. An adaptation of the method for determining the biological activity of rIL-7 using mononuclear cells of human peripheral blood was performed. The results of validation of this methodology based on indicators such as specificity, linearity, correctness and precision proved the possibility of using this method for routine analytical quality control of rIL-7 preparations.

The quality profile of nasal spray on the basis of rIL-7 was substantiated, which corresponds to the requirements of the guidance documents, the development of the formulations of the preparation and the technology for its manufacture was carried out. By determining the change in bioavailability of rIL-7 in vitro for 1 year (antiviral activity against HCV, HSV-2 and influenza virus, proliferation of the human MRCC), the composition of the drug that provides the best stability indicators (a prescription based on nipagine and a stabilizer PEG-400).

It is established that the developed drug possesses antimicrobial activity in relation to test microorganisms (*Bacillus subtilis*, *Candida albicans*). In order to neutralize antimicrobial activity, the effectiveness of using lecithin and polysorbate as antibacterial effect removing agents in combination with dilution of the test sample was proposed and tested 50 times when tested for its microbiological purity.

Gubina-Vakyulyk G.I., Sorokina I.V., Denisenko S.A.

**MICROSCOPIC FEATURES OF THYMUS IN WISTAR RATS AFTER THE
LONG-TERM INTAKE OF COLOR ADDITIVE TARTRAZINE (E102)**

Kharkiv National Medical University, Kharkiv, Ukraine

svetlanadeni@gmail.com

Food dye tartrazine (E102) is widely used in the food and pharmaceutical industries. Researches have demonstrated the development of metabolic disorders [B. Saxena, 2015] and nephropathies with elements of immune inflammation [G. Gubina-Vakyulyk et al 2017] due to its consumption. The study of immune shifts under the chronic oral administration of tartrazine is of particular interest.

The aim of our research was to study microscopic features of thymus in rats after the long-term tartrazine administration.

Materials and methods. Two-month-old Wistar rats (n=12) received 1 ml of 0.1% tartrazine solution per 100 mg of animal weight firstly daily intragastrically (using a probe) for 6 months and later as a component of diet. Thus, the intake was 7.5 mg / kg of body weight per day [A. Buldakov, 1996]. Rats from the control group (n=10) were kept under the same conditions and received physiological solution instead of tartrazine solution.

Animals were killed by decapitation. Their thymus was used for morphological investigation. Tissue was stained with hematoxylin-eosin and Einarson gallocyanin-chrome alum. Lymphocyte and macrophage morphometry and typing were performed. The "Graph Pad Prism 5" application was used for the statistical analysis.

Results and discussion. Hyperproliferation of the lymphoid component of the thymus with the formation of the so-called follicular hyperplasia was observed in the animals from the experimental group. The epithelial component of the thymus is characterized by a higher morphofunctional activity. The "starry sky" appearance in the cortex, as well as the presence of plasmocytes and plasmablasts in the follicles and perivascular spaces of the medullary area, was found.

The population of immature forms - pre-T-lymphocytes (Thy-1), B-lymphocytes (CD45RA) and macrophages (ED1) significantly increased among the thymocytes against the background of the decreased population of mature CD4 T-helpers and the trend to an increase in the number of CD8-T suppressors.

Conclusions. Animals that daily received the adequate dose of tartrazine for 6 months have microscopic signs of both cell-mediated and humoral immune response development in thymus.

Ivanivska T.S.¹, Shvets Y.V.²

**MORPHOCYTOCHEMICAL AND IMMUNOPHENOTYPIC FEATURES OF
BLOOD AND BONE MARROW CELLS IN PATIENTS WITH
MYELOYDYSPLASTIC SYNDROME**

¹RE Kavetsky Institute of Experimental Pathology, Oncology and Radiobiology of the NASU, Kyiv, Ukraine;

²Taras Shevchenko National University of Kyiv, Ukraine

tetyana.its@gmail.com

Myelodysplastic syndromes (MDS) represent a heterogeneous group of clonal diseases with underlying pathology of hematopoietic stem cells resulting in an ineffective hematopoiesis. The dysregulation of the immune system, in particular, the suppression of hematopoietic precursor cells is of high importance in MDS pathogenesis.

The purpose of the study is to evaluate the T- and B-cell immunity and to analyze morphocytochemical and immunocytochemical features of blast cells in patients with one of MDS forms (refractory anemia with excess blasts – RAEB).

Results. Blood and bone marrow preparations of 16 patients of the elderly with suspected MDS have been analyzed. In the hematopoietic cells of all patients, dysplastic changes were detected: hypogranular and hyper-segmental neutrophilic leukocytes, lobulated nuclei, and internuclear bridges in normoblasts, Jolly's bodies, and giant platelets.

The criteria for delineating RAEB-1 were up to 5% of blasts in peripheral blood and 5-9% of blast cells in the bone marrow. The diagnosis of RAEB-2 required 10-19% blasts in the bone marrow.

In both types of RAEB, the low-differentiated blasts with features of myeloid lineage were detected with weak diffuse acid phosphatase reaction; acid nonspecific esterase was negative. At least 3% of blast cells demonstrated positive myeloperoxidase activity. Expression of HLA-DR, CD34, CD33, CD117, CD13, CD15 was determined on a large part of the blasts.

Among lymphocytes, T cells were predominant ($CD3 = 64.8 \pm 5.7\%$, $CD16 = 10.2 \pm 2.7\%$, $CD19 = 10.1 \pm 1.6\%$). The ratio of T cell subpopulations and the T-helper/T suppressor index was within the normal range ($CD4 = 39.8 \pm 4.2\%$, $CD8 = 26.4 \pm 3.4\%$, $NKT\text{-cells} = 7.15 \pm 1.45\%$).

Conclusions. In patients with refractory anemia with excess blasts, the cellular arm of the immunity was not impaired. The dysplastic changes of all hemopoietic lineages with preserved myeloid commitment of blast differentiation have been identified.

Kopiika V.V., Bekasova O.F., Ivanova K.D.

PROGNOSTIC VALUE OF CYTOMORPHOMETRIC PARAMETERS OF
PERIPHERAL BLOOD LYMPHOCYTES IN THE DEFINITION OF OVARIAN
HYPERSTIMULATION SYNDROME

Zaporizhzhya National University, Zaporizhzhya, Ukraine

vkopijka@ukr.net

Ovarian hyperstimulation syndrome (OHSS) is a complication arising from exogenous gonadotropic therapy, which is used to obtain a large number of follicles in programs of assisted reproductive treatment. OHSS is characterized by a pronounced immune system reaction - proinflammatory processes are triggered with the corresponding activation of leukocytes.

The study was conducted in groups: 1) women who did not have a risk of developing OHSS (control group - CG); 2) at risk of developing OHSS; 3) women with clinical manifestations of hyperstimulation.

In groups at risk of OHSS and OHSS in relation to the control was observed moderate leukocytosis. The increase in the total content of leukocytes occurred against the background of physiological values of the level of C-reactive protein, which indicates an absence of inflammatory processes in the body of examined women.

In the study groups, multidirectional dynamics of functional cytomorphometric classes of lymphocytes was also observed: in CG and in women with a risk of OHSS, the majority of cells belonged to the average (CL 7-9 μm) and large size classes (CL $\geq 10,0$ μm). Another picture was observed in the group with OHSS - a significant increase in the content of small size classes of lymphocytes (CL $\leq 6,0$ μm).

Reduced proliferative capacity of cells (RBTL on PGA) in women at risk of OHSS and with clinical signs of OHSS is associated with the corresponding dynamics of cytomorphometric lymphocyte parameters: in the group with the risk of OHSS, the majority of lymphocytes belong to CL $\geq 10,0$ μm and they are already activated in peripheral circulation and belong to transitional forms or small blasts; in women with signs of OHSS, an increase in CL $\leq 6,0$ μm (post-proliferative differentiated lymphocytes with a high migration ability) occurs as a result of a

possible depletion of the $CL \geq 10,0 \mu m$, according to the pathogenetic approach to assessing the state of the immune system, is evidence of greater activation and tension of immunity in women with OHSS than with the risk of OHSS.

Thus, the cytomorphometric parameters of peripheral blood lymphocytes can be used as a prognostic criterion in determining the OHSS.

Kostyrko I.O., Yehorova S.U.

THE ROLE OF TH17 IN THE PATHOGENESIS OF SYSTEMIC AUTOIMMUNE DISEASES.

¹State Establishment «Dnipropetrovsk medical academy of Health Ministry of Ukraine», Dnipro, Ukraine.

gar4ild@gmail.com

Systemic autoimmune diseases are a global problem of modern society. This group of diseases is characterized by the loss of tolerance to the own antigens. A special role in the development of this group of diseases is played by Th17 and cytokines produced by these cells.

Th17 are derived from naive CD4+T cells in response to stimulation of IL-6, IL-23, IL-1 β and TGF- β . IL-6 and IL-23 activate STAT3, which increases the expression of the transcription factors ROR γ t and ROR γ , which in turn increase the expression of the main cytokines of this clone - IL-17A, IL-17F, IL-21 and IL-22. Th17 cells are important for protecting the host against extracellular pathogens, they play a key role in the activation of neutrophils. However, in infections caused by intracellular pathogens, activation of Th17 is pathological rather than protective. They inhibit the apoptosis of infected cells, contributing to the persistence of the infection. Induction of chronic inflammation and prolonged activation of Th17 by the pathogen leads to the development of immunopathological reactions of the organism and autoimmune tissue damage in the site of inflammation.

One of the most common autoimmune diseases is rheumatoid arthritis (RA). RA is a chronic autoimmune disease of the joints, characterized by inflammation of the synovial membrane and leading to destruction of the cartilage and bone. It has been established that Th17 cell products, such as IL-17 and IL-23, are present in the serum, synovial fluid and tissues of most patients with RA, but are absent in osteoarthritis and in healthy people. In the co-cultivation of peripheral T cells of patients with RA with synovial fibrocytes in the presence of type II collagen, the production of IL-15, TNF- α and IL-18 is stimulated by synovial cells. In response to induction, T cells produce high levels of IL-17 and INF- γ . IL-17

induces the secretion of IL-6, which causes destruction of the cartilage, inhibits collagen synthesis, and stimulates bone resorption in patients with RA.

These studies make it possible to improve understanding of the mechanisms of autoimmune diseases, which in the future will promote improvement of diagnostics and treatment.

Kotsyuba O., Lukianova N., Borikun T.

**PROGNOSTIC VALUE OF C-REACTIVE PROTEIN LEVEL IN BLOOD SERUM
OF BREAST CANCER PATIENTS**

RE Kavetsky Institute of Experimental Pathology, Oncology and Radiobiology, Kyiv, Ukraine
kotsyubaolha@gmail.com

Introduction. A necessary requirement for the appointment of adjuvant therapy in breast cancer therapy is the availability of information about the stage of the disease, presence of the metastases, and the molecular subtype of tumor. The prognosis of the cancer process requires more detailed evidence, as well as new informative markers.

Thus, our aim was to determine the relationship of serum C-reactive protein levels in breast cancer patients with the characteristics of the disease course.

Materials and methods. The study was conducted using 54 serum samples of BC patients with I-II disease stage, age ranged between 30 and 70 years. C-reactive protein levels were estimated using ELISA. We analyzed the relationships between the C-reactive protein level and clinical-pathological parameters. All measurements were performed in triplicate. Statistical analysis was performed using STATISTIKA 6.0 software.

Results. Analysis of serum C-reactive protein levels relation with the stage of breast cancer showed no correlation. However, it was found that the level of C-reactive protein in the serum is associated with the presence of the metastatic lesion in regional lymph nodes. In particular, in the serum of patients without metastases, the average level of C-reactive protein was 3.1 ± 0.7 mg/l, whereas, in patients with breast cancer of the N2-5 and N5-10, its levels were higher than 5.5 mg/L and in average counted 14.3 ± 3.5 mg/l. Correlation between the molecular subtype of breast cancer and the level of C-reactive protein in the blood serum of patients was not detected.

Conclusion. Obtained data are the groundwork for further exploration of C-reactive protein in the blood serum of patients as the marker of the breast cancer metastasis and for estimation of breast cancer course.

Kutasevich Ya. F.¹, Gubina-Vakyulyk G.I.², Bronova I.M.¹, Gorbach T.V.²
**CD4 AND CD8 LYMPHOCYTES IN NONPURULENT LEUKOCYTIC DERMAL
INFILTRATE IN ACNE DURING PATHOGENETIC THERAPY**

¹U "Institute of Dermatology and Venereology of the National Academy of Medical Sciences of Ukraine";

²Kharkiv National Medical University, Kharkiv, Ukraine

GVGIpatology@yandex.ru

Acne is a chronic recurrent dermatosis with multifaceted pathogenesis that occurs in adolescents, males and females of reproductive age. Using biopsy material, it has been established that the formation of a focus of purulent inflammation in the area of the hair follicle is preceded by the appearance of a macrophage-lymphocytic infiltrate located around the epithelial structures of the skin.

The **aim** is our research was to study the presence of CD4 and CD8 cells in a dermal macrophage-lymphocyte infiltrate in patients with severe acne.

Materials and methods. In patients with severe acne after their examination and exclusion of contraindications, biopsy of the skin from the interscapular region was performed prior to the prescription of treatment with systemic retinoids (0.4 mg per kg of body weight daily for 5-6 months), after 6 weeks, and after 4-6 months after the beginning of treatment (12 biopsies). Immunohistochemical investigation was carried out using antibodies to T helper (CD4) and T suppressor / cytotoxic lymphocyte (CD8) antigens (TermoScientific, USA).

Results. Prior to treatment, CD8 lymphocytes were numerous in macrophage-lymphocytic infiltrates, whereas CD4 lymphocytes were rare. During the treatment, there was an increase in the number of CD4 lymphocytes and a decrease in the number of CD8 lymphocytes. Clinical convalescence was accompanied by the disappearance of dermal leukocytic infiltrates, CD8 and CD4 lymphocytes were rare. We have presumed that CD8 lymphocytes prevailed before treatment are cytotoxic. In blood of such patients an increase in the absolute and relative amount of cytotoxic T lymphocytes has also been revealed (AV Kormilitsina, NM Kalinina, 2017).

Conclusions. There are elements of immune inflammation in the skin in the morphogenesis of severe acne. Systemic retinoids used to treat acne cause elimination of inflammation.

Lahutina O.S.**EVALUATION OF THE INFLUENCE OF IRON OXIDE NANOPARTICLES ON
NATURAL IMMUNITY AND IMMUNE ORGANS OF RATS**

SI" Kundiiiev institute of occupational health of the National academy of medical sciences of Ukraine", Kyiv, Ukraine

lagutinao@ukr.net

Iron oxide nanoparticles (NP) are used in medicine and biology. In most cases, their using involves entering to the human body, therefore, requires safety assessment.

The aim of this work was to investigate the effect of Fe_2O_3 nanoparticles of 19 nm, 75 nm and 400 nm on rats natural immunity in conditions of 30-fold intraperitoneal administration into the body.

During the study, the iron content in the blood and immune organs of the rats were measured by atomic emission spectroscopy, complete blood count with white blood cell count. Natural immunity indicators were determined by phagocytic and bactericidal activity of peritoneal macrophages and neutrophils, content of zinc protoporphyrin, albumin, globulin and circulating immune complexes, complement activity in serum. Evaluated structural changes in the thymus and spleen, as well as the accumulation of iron, staining with hematoxylin-eosin and the Pearl reaction.

It was investigated that injections of Fe_2O_3 nanoparticles within 30 days in a dose of 0.001 mol / L caused an enhancement of iron level in the blood and immune organs, indicating active movement of nanoparticles by blood flow and lymph flow, accumulation in organs.

In experimental rats, compared with control rats, hemoglobin levels were decreased, zinc protoporphyrin levels were increased, leukemia and lymphopenia were moderately expressed, and monocytes and eosinophils amount was increased. Fe_2O_3 NP stimulated phagocytic activity and respiratory burst in the neutrophils and peritoneal macrophages, reduced globulin level, and increased circulating immune complexes in serum.

In the thymus of rats were present large lymphocytes and plasmocytes; in the spleen was observed hyperplasia of white and red pulp, increased lymphatic follicles size and number of macrophages filled with iron, indicating the activation of the immune response in these organs.

Those changes are signs of anemia development, activation of the cellular and humoral link of natural immunity and response in the immune organs, which were more expressed after injections of Fe_2O_3 NP with size 19 nm.

Lazarenko L.M.¹, Melnykova O.I.^{1,2}, Babenko L.P.¹, Falaleeva T.M.², Spivak M.Ya.¹

LACTIC ACID BACTERIA AND BIFIDOBACTERIA AFFECT THE METABOLISM OF LIPIDS AND CARBOHYDRATES AND CYTOKINES PRODUCTION ON THE EXPERIMENTAL MODEL OF OBESITY

¹D.K. Zabolotny Institute of Microbiology and Virology of NASU, Kyiv, Ukraine;

²Taras Shevchenko National University of Kyiv, Ukraine

alex190697@ukr.net

The metabolic disturbances, systemic and vascular chronic low-grade inflammation as well as the disbalance of the gut microbiota may be observed in the case of obesity that can cause a number of human's diseases. It has been shown that some probiotic strains of lactic acid bacteria (LAB) and bifidobacteria are able to improve these important biomarkers of the obesity.

The aim of the work was to determine the possibilities of normalizing the lipids` and carbohydrates` metabolism and the proinflammatory cytokines production by the using of probiotic strains – *Lactobacillus casei* IMV B-7280, *L. delbrueckii* subsp. *bulgaricus* IMV B-7281, *Bifidobacterium animalis* VKB, *B. animalis* VKL (individually) or *L. casei* IMV B-7280 / *B. animalis* VKB / *B. animalis* VKL and *B. animalis* VKB / *B. animalis* VKL compositions in case of experimental obesity.

To simulate obesity, BALB/c mice (6-8 weeks) obtained the fat-enriched diet (fats – 30 %, proteins – 40 % and carbohydrates – 30 %). Obese mice received standard feed and a suspension of probiotic bacteria. The levels of cholesterol and carbohydrates as well as tumor necrosis factors- α (TNF α) in serum blood were determined by standard biochemical methods and enzyme immunoassay respectively. The analysis of gut microbiota was carried out using standard microbiological methods.

It was found that these probiotic strains of LAB and bifidobacteria and probiotic compositions reduced the weight of obese mice that was associated with decrease the level of cholesterol, glucose and TNF- α in serum blood. Partial normalization of gut microbiota by reducing the number of opportunistic microorganisms and increasing the number of LAB and bifidobacteria was also observed in probiotic treated obese mice. *L. casei* IMV B-7280 and *L. casei* IMV B-7280 / *B. animalis* VKB / *B. animalis* VKL composition were more effective.

So, *L. casei* IMV B-7280 and *L. casei* IMV B-7280 / *B. animalis* VKB / *B. animalis* VKL composition are recommended for creation of effective probiotic drugs directed to prevent metabolic disturbances, systemic chronic low-grade

inflammation as well as improve microecological malfunctions in gut that can be used to treat obesity.

Lisyanii N.I., Gnedkova I.A., Stanetskay D.N., Lisyanii A.A., Belskay L.N., Potapova A.G.

TWO-FACED ROLE OF NEUTROPHILS IN ANTI-TUMOR IMMUNITY

The State Institution Romodanov Neurosurgery Institute, National Academy of Medical Sciences of Ukraine, Kyiv, Ukraine

nimun.neuro@gmail.com

Neutrophils belong to short-lived cells of innate immune system. These cells possess high migratory, phagocytic and cytotoxic activity, that allows them to provide anti-infective protection of the body. Neutrophils have the ability to accumulate both in the nidus of infection and in inflammatory focus as well as in tumor tissue, where they can exert immunosuppressive, tumor-stimulating action. These tumor-associated neutrophils (TAN) acquire a protumoral "N2" phenotype. TAN identification was among the first evidences of the existence of different neutrophil subsets: pro-inflammatory N1 and anti-inflammatory immunosuppressive N2. The mechanisms for these neutrophil phenotypes formation are just beginning to be elucidated. However, growing number of clinical observations shows that neutrophilia is associated with poor prognosis in several types of cancers. The aim of this work was to study the proportion of neutrophils and other immune cells in peripheral blood of patients with brain tumors of different histology type and different malignancy grade. Results of our investigation revealed the dependency of circulating neutrophil fraction from tumor histotype and malignancy grade. It was found that highest neutrophil fraction was in patients with neuroectodermal and mesenchymal tumors, and the smallest - with neuroectodermal tumors. The ratio of the fraction of CD16+ neutrophils (cells of innate immunity) to lymphocytes (cells of adaptive immunity), especially CD4+ cells, was greatest in patients with neuroectodermal tumors. It indicates an imbalance in the activation of cells of these parts of the immune system: augmentation of non-specific inflammatory component along with down-regulation of specific T-cell-mediated immunity. Therefore, comparing our results with the literature data, one can conclude that the growth of malignant brain tumors, unlike the growth of benign tumors, is accompanied by both an increase in the fraction of circulating neutrophils and by the alteration in the phenotype of their counterparts which are recruited to the tumor tissue.

Lisyanii N.I.¹, Pilipchuk V.C.¹, Belskay L.N.¹, Lisyanaya T.A.², Ponomaryova I.G.²

**THE EFFECT OF THE PREPARATION «DZHERELO-1» ON THE IMMUNITY
AND ANTIINFECTIVE RESISTANCE**

¹The State Institution Romodanov Neurosurgery Institute, National Academy of Medical Sciences of Ukraine, Kyiv, Ukraine;

²The State Institution PAG Institute, National Academy of Medical Sciences of Ukraine, Kyiv, Ukraine

nimun.neuro@gmail.com

Various groups of immunomodulating drugs have been widely used in the modern clinical immunology, namely, glucocorticoids, monoclonal antibodies, interferons, adjuvants. At the same time, there is a limited list of drugs that have a mild therapeutic and preventive effect on the immune system. The purpose of our research was to study the effect of the complex herbal preparation "Dzherelo-1" on the local immune system and microbiocenosis of the oral cavity, namely, on microbial and viral contamination of the oral fluid. The drug "Dzherelo-1" is a complex alcohol-water extract of 12 plants with immunomodulating and antioxidant properties. The drug is applied in the form of drops for 14-16 days (according to the instructions) and is recommended for healthy people. Investigation of immunological, microbiological and virological parameters of the oral fluid was carried out before the beginning of the drug and on 2,16,45 days after the end of the course. It was established a 3-4-fold increase in the level of α -interferon in saliva immediately after the end of the drug administration, the level of sIgA increased during the period from 15 to 45 days, that indicates activation of local adaptive immunity. The total microbial number in the oral fluid was decreased from $10^{8.4}$ cfu / ml to $10^{5.4}$ cfu / ml on day 30 of the drug administration. The number of fungi, gram-negative pathogens, hemolytic staphylococci was significantly lower than that before the drug administration. Leukocyte fraction and the number of epithelial cells with adhesive microbial associations was also decreased in oral fluid. The detection of herpes viruses (HV) in the oral fluid by PCR method revealed only HV 4,6 and 7 in 85-90% of cases. HV 1, 2 and CMV viruses were not detected. In the early period after the application of the drug "Dzherelo-1" there was a decrease in the DNA level of HV4 and 6. The content of DNA of HV7 was reduced only on the day 45 of observation period. Our results confirm the immunomodulating and anti-infective characteristics of the "Dzherelo-1", and allow to recommend it as curative and preventive drug to activate local immunity and to increase resistance to infectious agents.

Matvyeyeva S.L., Shevchenko O.S., Stepanenko A.L.

**MODULATION OF T-CELL MEDIATED IMMUNITY BY THYROID FUNCTION IN
TUBERCULOSIS PATIENTS**

Kharkiv National Medical University, Ukraine

Kjara.clair@gmail.com

The goal of study is to determine the influence of previous thyroid state on T-cell mediated immunity and chemotherapy response in patients with new case of pulmonary tuberculosis. Materials and methods: The parameters of thyroid state: free thyroxine (T4f) and thyroid stimulating hormone (TSH) and T-cell mediated immunity: tumor necrosis factor, interferon- γ and interleukins-2,-4,-6 were measured by ELISA in the serum of 2 groups of new cases of tuberculosis: main group of 54 patients with thyroid pathology found by ultrasound screening and control group of 46 with normal echostructure of thyroid. Results: In the group with thyroid pathology significant T4 level decreasing from (10.02 ± 0.16) pmol/ml to (12.24 ± 0.13) pmol/ml and TSH level increasing from (2.72 ± 1.31) IU/l to (1.37 ± 0.16) IU/l comparing with the control group were demonstrated. The levels of pro-inflammation cytokines: tumor necrosis factor, interferon- γ and interleukins-2,-6 in patients with thyroid pathology were significantly lower (responsively 60.84 ± 25.01 ; 3.74 ± 2.45 ; 7.08 ± 1.97 ; 51.87 ± 33.54) in compare with tuberculosis patient without thyroid pathology (responsively 30.77 ± 16.77 ; 1.22 ± 0.81 ; 4.88 ± 1.05 ; 16.98 ± 11.81). The level of anti-inflammation cytokine – interleukin-4 was significantly lower in control group (0.002 ± 0.003) in compare with group of thyroid pathology (0.030 ± 0.027) . Thus, thyroid pathology was associated with depressed cytokines response to tuberculosis. Chemotherapy efficacy estimated by traditional criteria used in phthysiology in tuberculosis patients without thyroid pathology was better than in tuberculosis patients with thyroid pathology. Conclusion made that screening of thyroid state is needed in new cases of tuberculosis to diagnose and treat cryptic thyroid pathology for reconstitution of immunity response and improving the results of antituberculosis chemotherapy.

Melnyk O.V.

**EVALUATION OF THE STATE OF THE CELL LINE OF IMMUNITY IN
PATIENTS WITH JOINTS INFLAMMATORY PATHOLOGY**

Danylo Galytsky Lviv National Medical University, Ukraine

viruszet8@gmail.com

Actuality. In recent years, an increase in the number of patients with inflammatory pathology of the joints has been observed. In the structure of these

diseases, rheumatoid arthritis RA and reactive arthritis ReA play an important role. Until now, a specific etiological factor has not been identified, which leads to their development. However, it is assumed that trigger factors may be not only microorganisms, but also their condition (fragments), causing infectious diseases, stresses, traumas, overcooling, hormonal factors, etc. The purpose of this work was to finding out the functional activity of cells of the phagocytic system and to identify the diagnostic value of phagocytic reactions in patients with inflammatory pathology of the joints. The results of the research showed that there is a high number of leukocytes in patients with RA and ReA 10,20g/l and 9,10g/l compared with healthy subjects 7.21g/l. The analysis of the number of lymphocytes shows that their absolute number CD45 in RA and ReA does not differ from the control values, but the relative amount in RA 21,94% is lower than the control values 34,53%. Concerning T-lymphocytes CD3, their absolute values for RA and ReA do not differ from the control, but at the same time, the relative values of this indicator at RA 68.80% and ReA 68.81% were lower than the control values 75.77%. Relative number of CD4+ lymphocytes in patients with RA and ReA was higher 45.11% and 45.20% relative to control values 34.23%. The absolute amount of CD4+ lymphocytes in patients with RA did not differ from the control, and in patients with ReA was significantly higher 1.2g/l from the control values 0.63g/l. The relative number of T-cytotoxic lymphocytes CD3 CD8 in patients with ReA was higher 25.90% relative to the control group 22.15%. The absolute and relative number of natural killer cells CD16 CD56 in patients with RA was higher 14.9% than in healthy subjects 9.96%. The growth of the immunoregulatory index value in RA against the background of a reduced amount of T-cytotoxic CD8 lymphocytes may indicate activation of the cellular link of autoaggression with the inclusion of regulatory mechanisms.

**Moskvina M.¹, Nesyn D.¹, Gorbach O.², Skackova O.², Inomistova M.^{1,2},
Khranovska N.²**

IMMUNOMODULATORY EFFECT OF DC VACCINE AND CISPLATIN IN LOW DOSES IN MOUSE SARCOMA 37 MODEL

¹Taras Shevchenko National University of Kyiv, Ukraine;

²National Cancer Institute of Ukraine

moskvina.maryna@gmail.com

Introduction. The lack of effective methods of cancer immunotherapy forces the search of new approaches for decreasing tumor immunosuppression. It is

known that the usage of chemotherapeutic drugs in low doses reduces the number of Treg cells.

Materials and methods. Fifty CBA mice were used in the experiment. Mice were injected with S-37 cells intramuscularly. Cisplatin was injected intraperitoneally 5 times in doses 0.2 mg/kg or 2 mg/kg on the 7th day after tumor transplantation at intervals of 1 or 3 days, respectively. Vaccine based on dendritic cells (DC vaccine) was injected intravenously into orbital sinus in concentration of 0.2×10^6 DC per animal 3 times on the 4th day after last injection of cisplatin at intervals of 3 days.

Results. Maximal immunomodulatory effect was observed at the combined scheme with 2 mg/kg dose of cisplatin and DC vaccine (CIT2). The number of CD4+CD25^{high} cells decreased after administration of DC vaccine and cisplatin 2 mg/kg as compared to the control group ($p < 0.05$). Moreover, the number of CD69+ and CD54+ cells in this group increased. CIT2 scheme application led to the increase of phagocytosis in splenocytes and peritoneal macrophages as compared to the control group (by 52.33% for neutrophils, $p = 0.04$; by 28.33% for spleen macrophages, $p = 0.028$; by 68.67% for peritoneal macrophages, $p = 0.025$). Insignificant increase was observed in the group of DC vaccine and cisplatin 0.2 mg/kg (CIT1). In CIT2 group we observed the growth of ROS stimulation coefficient by splenocytes and peritoneal macrophages comparing to the control group, $p < 0.05$. Allogeneic cytotoxic activity in splenocytes reached 37.4% in CIT2 group compared to the control group, $p = 0.011$. The application of CIT1 regimen led to the increase of syngeneic cytotoxic activity to 43.8% compared to the control group, $p = 0.035$.

Conclusion. The administration of cisplatin in low doses significantly reduces tumor immunosuppression and increases the DC vaccine efficacy.

Motorna N.V., Gumenyuk A.V., Sokurenko L.M., Savosko S.I., Chaikovsky Yu.B.

*HERPES SIMPLEX VIRUS 1 INFECTION INDUCES MORPHOLOGICAL
CHANGES IN BRAIN AND LIVER IN BALB/C MICE*

Bogomolets National Medical University

natalivfrcbv@gmail.com

Introduction. *Herpes simplex virus* type I (HSV-I) is quite prevalent in general population. HSV-I exists in latent form in the nervous system. The damage for other organs, particularly liver, is not studied enough. The aim of the current work was to study morphological features of systemic organs injury caused by HSV-I.

Materials and methods. Experiments were conducted on BALB/c line mice weighing 18-20 g. The animals were infected with mouse-adapted HSV-I in virological laboratory in the Gromashevsky L.V. Institute of Epidemiology and Infection Diseases of NAMS of Ukraine. On day 30 histological studies of the brain and liver were conducted. The virus in blood serum, brain and liver was assessed by PCR and dot-ELISA.

Results. Using PCR, dot-ELISA and histological methods the presence of HSV-I and organ damage was confirmed in 100% of samples. Focal infiltration of lymphocytes and monocytes in mice brain was observed in the corpus callosum, brain cortex and hippocampus. The inflammation caused neurodegenerative changes leading to reduction of neurons in the studied areas. Herpes infection in liver was marked by hyperemia of hemocapillaries and central veins and local hemorrhages. Some portal tracts demonstrated focal accumulation of neutrophils and monocytes. Cytopathological changes (cell dystrophy, hypertrophic nuclei) of hepatocytes were focal or diffuse. The patterns of herpetic liver infection are characterized by the absence of tissue basophils response and their absence in the stroma as well as by the delayed process of fibrosis.

Conclusions. The experimental data showed that HSV-infection is not limited to the neurodegenerative changes in the nervous system and can cause systemic damage of organs. Our study demonstrated features of systemic organ damage due to HSV-I viremia. These data also suggest spread and penetration of infection into the organs following viral damage of the blood vessel wall.

Nikolaenko D., Tymoshenko A.

ADAPTIVE HYPOTHESIS OF CONVERSION PrPc INTO PrPSc

Kiev International University, Kiev, Ukraine

profdmityrnikolaenko@gmail.com

Transmissible spongiform encephalopathies (TSEs) are a family of rare progressive neurodegenerative disorders that affect both humans and animals. It remains unexplained from the scientific standpoint. There are three hypotheses: "pure protein", multicomponent and viral. An adaptive hypothesis has been developed. The logic of adaptive hypothesis is as follows: 1. There are primary and secondary (transmission) prion contamination. 2. To understanding the process of transformation of PrPc into PrPSc, the study of soil protozoa is of fundamental importance. They and their ecology contain key information for understanding prion infectious diseases. 3. Probably the main importance in manifestation of pathogenic properties is the endosymbiosis of protozoa with

microorganisms. 4. Apoptosis is the main subject of research. There is a failure in natural manifestation of apoptosis. Starts embodiment, which can be defined as Q-apoptosis. 5. Q-apoptosis is triggered as a consequence of an external signal. For example, a certain change in the microelement and electromagnetic characteristics of a habitual microorganism. 6. Consequence of Q-apoptosis is manifestation of some prion infectious diseases. They can be both single and massive. 7. The brain is an aggressive microelement and electromagnetic environment for normal protein-protein interactions. Provided prions get into the brain, the adaptation process begins. A comfortable microelement and electromagnetic environment is formed. Prions perceive reality through the prism of the microelement and electromagnetic fields. 8. Starts TSEs. The formation of cavities in brain cells, neuronal degeneration, proliferation of connective tissues in place of dead nerve cells, the formation of amyloid plaques formed from clusters of pathological prions, atrophy of the warm-blooded brain. This is a manifestation of the adaptation process. 9. This occurs in the absence of inflammatory reactions of the body. It is not an infection in its traditional sense. It is an infection - a transformation. 10. Experimental work can be carried out. As a model disease, scrapie can be taken.

Novosad N.V., Ivanova A.V.

**LEUKOGRAM OF BLOOD AND METABOLIC ACTIVITY OF NEUTROPHILIC
LEUKOCYTES IN PRESCHOOL CHILDREN WITH COMMUNITY-ACQUIRED
PNEUMONIA**

Zaporizhzhya National University, Zaporizhzhya, Ukraine
novosadnata@gmail.com

Over the past few years pneumonia has been occupying one of the leading places in the structure of bronchopulmonary pathology in patients of childhood. Pathogenetic disorders in community-acquired pneumonia (CAP) are closely connected with the features of neutrophilic granulocytes and changes their metabolic state determines the development and effects of acute pneumonia. Myeloperoxidase (MPO) and non-enzyme cationic proteins (CP) are the main factors of bactericidal activity phagocytes.

In preschool children with CAP were studied leukogram of blood, activity of MPO and level of CP in neutrophilic leukocytes and the amount of MPO and CP-positive neutrophils.

As the results of the research showed in sick children the total number of leukocytes increased by 1,7 times, which by 22% exceeded the upper limit of the

physiological norm, also elevated in 2,4 times the relative number of band neutrophils and by 23% the number of segmented neutrophils; in 1,7 times decreased the number of lymphocytes. Activity of MPO and level of CP in sick children were 15% higher than in healthy patients. The weakly positive reaction of MPO in the form of small scattered granules was observed in 12,45% of cells and did not differ significantly from the control indicator. In 3,2 times increased the number of neutrophils with a sharp positive reaction and in 2,5 times decreased the number of neutrophils with moderate content of granules. Similar changes were observed in the values of CP-positive cells.

In the analysis were detected various atypical changes in neutrophils of blood. There was a toxic granulation in cells, which often accompanied by the formation of cytoplasmic vacuoles, increased the number of hypersegmented neutrophils, and also detected karyorrhexis,

Thus, in children with CAP, when increasing the relative number of neutrophilic leukocytes, increases the activity of the MPO and the level of CP; elevates the number of neutrophils with atypical changes and with a strongly positive reaction of MPO and CP, indicating an increase the number of primed neutrophils compared with the conventionally healthy group.

Oliynyk D.M.¹, Chambers B.J.²

THE INVESTIGATION OF PD-1 EXPRESSION ON NK CELLS

¹National University, Kyiv, Ukraine;

²Karolinska Institutet, Stockholm, Sweden

oleynikdenis3007@gmail.com

Natural killer (NK) cells are an important part of innate immune response, since through the production of cytokines and cytotoxicity they can deal with transformed and stressed cells. However, tumor cells often evade the NK cells' defense line using PD-1:PD-L1 pathway. PD-1 is a member of Ig family, which is located on a cell surface playing an important role in down-regulating the immune system and promoting self tolerance by inhibiting T cell functions. In present study, we investigated the expression of PD-1 on subsets of mouse and human NK cells.

The aim of the study was to prove the suggestion that the mix of IL-12/IL-15/IL-18 could induce PD-1 on both human and mouse NK cells, to find the leading subsets in PD-1 expression and investigate if PD-1 and PD-L1 can create a cis-interaction on the NK cell's surface.

Mouse NK cells were obtained from either frozen or fresh spleen of B6 mice. PD-1 KO NK cells were obtained from *Pdcd1*^{-/-} mice, seeded at 1×10^6 cells/mL. Human NK cells were obtained from PBMC. NK cells were counted and seeded at 3×10^6 cells/mL. After culturing, purified NK cells were processed for immunofluorescence staining. After the staining cells were measured on a BD LSR II flow cytometer. Fluorescence correlation spectroscopy measurements for the investigation of PD-1/PD-L1 interaction was performed using a Zeiss 510 microscope.

We now can be sure that cytokine stimulation with IL-15/IL-12/IL-18 could induce PD-1 on both human and mouse NK cells. Obtained data suggests that KLRG1⁺ among mouse and CD56^{dim}CD16^{dim/neg} among human NK cells seems to be the leading subsets in PD-1 expression. In addition, we investigated if PD-1 and PD-L1 can create a cis-interaction on the cell's surface. According to the fluorescence correlation spectroscopy data, PD-1 might create a cis-interaction with PD-L1 but certain results are still unclear. Preliminary data showed the lower speed of PD-L1 on PD-1 KO mice line than on B6 mice NK cells. This suggests that, if PD-1:PD-L1 cis-interaction exists, it can be useful for further investigations in self-regulation of NK cells and in modulation of cancer pathways.

Oliynyk N.M

IMMUNE STATUS IN THE CONTEXT OF A HEALTHY LIFESTYLE

I. Horbachevsky Ternopil State Medical University, Ternopil, Ukraine

oliynyknimy@tdmu.edu.ua

The immune system is one of the structures that performs an integrating as well as corrective function in maintaining human health.

According to World Health Organization, human health is 50% dependent on lifestyle, and only 10% on the quality of care. However, there is no doubt that the state of the immune system and its operation is almost entirely dependent on lifestyle. In particular, physical activity is considered as a nonspecific activator of immunity. The first to exercise are peripheral blood cells - neutrophils, which provide not only phagocytosis of bacteria and viruses, but also the synthesis of immunoregulatory factors. This phenomenon is termed "leukocytosis of physical exercises". The total effect of physical exertion is explained by both direct and indirect (via hemostasis, nervous, cardiovascular, endocrine) effects on general and local immunity.

The immunomodulatory effect of rational nutrition is associated with its effect on the most parts of the immune system. Deficiency of the main antioxidant

nutritional factors, for example, selenium, vitamin E, significantly disturbs the normal maturation of the population of immunocompetent cells. Vitamin E increases the activity of natural killers, stimulates phagocytosis as well as regulates the thymus gland. Reducing the reactivity of cellular immunity, the production of antibodies and antitumor activity is observed when retinol is deficient. Vitamin C stimulates macrophages, increases the production of interferons, increases antiviral activity, activates complement, promotes the synthesis of IgM, IgA. It is known that during a prolonged stress, with a sense of anxiety, the mass of immunocompetent organs (thymus, spleen, lymph nodes) decreases, the ratio between the subpopulations of lymphocytes changes, and the production of cytokines of the acute phase of inflammation increases. Low control over the stressful situation leads to a decrease of the T-helpers, and in individuals with high stress control, the number of the T-lymphocytes increases.

Therefore, the criteria of a healthy lifestyle - directly affect to the state of adaptive immunity and are sufficiently effective soft immunomodulators.

Oriabinska L.¹, Lazarenko L.², Belur D. Prasanna³

**IMMUNOMODULATORY PROPERTIES OF TANNASE POSITIVE STRAIN
LACTOBACILLUS PLANTARUM 2621**

¹National Technical University of Ukraine "Igor Sikorsky Kyiv Polytechnic Institute", Ukraine;

²Institute of Microbiology and Virology. D.K. Zabolotnoho, Kyiv ;

³National Institute of Technology Karnataka, Surathkal, Mangalore, India

olanab9@gmail.com

The possibility of creating functional products based on lactic acid bacteria with antioxidant activity is a matter of great interest. One of the criteria for selection of probiotic strains for functional food is their moderate immunological reactivity. With this purpose immunomodulatory properties of the *L. plantarum* 2621 which has a tannase activity were studied. The functional activity of cells was evaluated by using conventional methods. The study was conducted on intact mice of line BALB/c, female, age 8 weeks, that intragastrically received a lactic acid bacteria once a day during 7 days with concentration of $1 \cdot 10^6$ cells/ml. On 1st and 9th days after administration of bacteria to mice PhN, IPh and indicators of spontaneous macrophages NBT-test increased slightly, but the difference in comparison with the control was not significant. In the mice spleen the numbers of CD3 +, CD4 +, CD8 + cells were the same as in control throughout the observation period. There was a trend of immunoregulatory index CD4 / CD8 increasing for 1 night, and the number of CD19 + cells in the spleen on day 9th,

but the difference in comparison with the control was also not significant. In order to assess of probiotic strain changes in cytokine production, such as IFN- γ , IFN- α , IL-17A, IL-23 studies have shown that in 3 and 6 days after administration of the probiotic suspension to intact mice, an increase in the content of IFN- α in serum was observed. This may indicate the possibility of activation under its influence as nonspecific antiviral defense of the organism, as well as innate immunity. Instead, the production of Th1-type cytokine-IFN- γ in mice in this group and IL-23 and IL-17A did not change.

Thus, in the absence of apparent deviations in immunological parameters from the control group, it has been established that the probiotic strain *L. plantarum* 2621 does not exhibit general immunomodulatory activity, but exhibits non-specific antiviral activity. This fact allows us to consider *L. plantarum* 2621 as a promising producer for the production of functional food.

Osyphchuk D.V.^{1,2}, Chernyshov V.P.¹, Gilfanova A.M.², Bondarenko A.V.²

**FUNCTIONAL TESTS, AS A PART OF IMMUNE TESTING, FOR IN-DEPTH
ASSESSMENT THE CURRENT STATUS OF THE IMMUNE SYSTEM IN
CHILDREN**

¹Institute of Pediatrics, Obstetrics and Gynecology, Kyiv, Ukraine;

²Shupyk National Medical Academy of Postgraduate Education, Kyiv, Ukraine
dariia_osyphchuk@ukr.net

Problem Measurement of "immunity" has often focused on measurement of humoral immunity to detect the presence of protective antibodies and on quantitative assessment of cellular immunity. However, such assays not always enough to find out the cause of impairment immune function. Functional assays for evaluation of cellular immune response in vitro are more complex and less easy to measure, but could measure current cellular immune assays response in vitro, by elicitation of a functional response at the time of the test.

Method: a whole blood assay to evaluate activation response of immune cells to toll-like receptors (TLR) agonists was developed. Heparin-treated blood was sampling from 18 healthy control subjects and from 25 patients (study group) with recurrent respiratory infections with accompanied by bacterial complications (otitis, pneumonia, sinusitis). Previously, all patients from the study group were tested with the screening immune tests and no abnormalities were detected.

Results: Stimulation of TLR2, TLR4, TLR7 and TLR8 led to a significant increase in the expression of integrin CD11b on granulocytes. The level of

expression of CD11b after stimulation of TLR7 / 8, was lower in the study group - $383,2 \pm 95,8$ MFI, compared with control group - $557,7 \pm 172,6$ MFI, $P < 0.05$.

Also, we examined level of CD69 (marker of activation) expression on NK - lymphocytes after incubation with TLR3 ligand in both groups. There was a significant decrease in the level of an activation marker – CD69 on NK cells $36,3 \pm 4,4\%$ in the group of children with recurrent infections, compared with a control group of healthy children ($56,5 \pm 4,9\%$). Such decreased expression of adhesion molecules on granulocytes in response to TLR stimulation accompanied by reduced activation of NK-lymphocytes, can lead to disruption of migration to the site of infection and promote increased sensitivity to viral infections and bacterial complications.

Conclusion: developed assay can be used as a part of immune testing to evaluate a patient with recurrent infections with no detectable abnormality of antibody function, complement activity, neutrophil function, or cell mediated immunity.

Petishkina V.M.¹, Skivka L.M.², Koposova I.V.¹, Firsova A.S.¹

**FEATURES OF GENERATION OF ACTIVE FORMS OF OXYGEN BY
MONOCYTES OF PERIPHERAL BLOOD OF PATIENTS WITH COPD
EXACERBATION**

¹SO "National institute of phthisiology and pulmonology named after F.G. Yanovskiy NAMS of Ukraine";

²Taras Shevchenko National University of Kyiv, Kyiv, Ukraine
koposova@ifp.kiev.ua

The investigation of the generation of active forms of oxygen by monocytes of peripheral blood was held among patients with COPD exacerbation.

The objective of the work was to investigate the generation of active forms of oxygen by monocytes of peripheral blood in 18 COPD patients and 30 blood donors using the method of flow cytofluorometry.

There was found out the reduction of ROS production in DCFDA-marked white blood cells of whole blood: monocytes - ($20,7 \pm 1,7$ standard units) in comparison with the control ($36,6 \pm 4,2$ standard units) $p < 0,01$. The opsonized zymosan was used for the measuring of activity of process of ROS creation under the effect of inhibitors of oxygen-dependent metabolism. There was found out the increasing of level of stimulated ROS production by monocytes ($227,1 \pm 21,6$ standard units) in comparison with monitoring indicators ($165,7 \pm 9,2$) $p < 0,02$, the coefficient of stimulation in patients was higher ($13,1 \pm 1,7$) than the indicators of control group ($7,0 \pm 0,85$) $p < 0,01$.

Thus, the ROS production by monocytes of peripheral blood is characterized by reducing the intensity of fluorescence without stimulation, though under the effect of zymosan there was found out the increasing of this indicator, which allowed to assess the functional reserve of cells in response to the stimulation in COPD patients.

Popyk A.¹, Tiuliukin I.², Zantaraia T.², Zub O.³, Susak Y².

**THE EFFECT OF ULINASTATIN ON OXIDATIVE METABOLISM AND
PHAGOCYTIC ACTIVITY OF CIRCULATING LEUKOCYTES IN PATIENTS
WITH SEVERE ACUTE PANCREATITIS**

¹Taras Shevchenko national university of Kyiv, Ukraine;

²Department of surgery, O.O. Bogomolet's National Medical University, Ukraine;

³Department of surgery N2, City Clinical Emergency Hospital of Kyiv, Ukraine

angelina230395@gmail.com

Background: Leukocytes are central players in acute pancreatitic (AP) pathogenesis, orchestrating the initiation, propagation, as well as local and systemic complications of the disease. Ulinastatin (Ust) is a serine protease inhibitor, that exert anti-inflammatory and anti-oxidant effects by attenuating the TLR/NF- κ B pathway activation. Ust is successfully used in the treatment of AP due to the ability to inhibit effectively the proteolytic enzymes and to attenuate systemic inflammatory response. This study was aimed to investigate the effect of Ust on functional state of circulating leukocytes in patients with severe acute pancreatitis (SAP). **Methods:** 22 patients with AP were enrolled in the study. Oxidative metabolism and phagocytosis activity of circulating leukocytes was examined by flow cytometry. Reactivity reserve of investigated functions was evaluated after the treatment of blood samples with phorbol 12-myristate 13-acetate in vitro. **Results:** Both standard treatment and therapy with Ust were associated with the decrease reactive oxygen species (ROS) generation along with the increase of their endocytosis activity in circulating leukocytes without restoring the metabolic reserve of these cell function. The investigated indices of circulating leukocyte metabolic activity after the treatment with Ust in patients with AP were characterized by significant individual variability, that led to a lack of statistical reliability of the revealed differences between the treated and control groups. Patients can be divided in two subgroups with different metabolic state of circulating phagocytes after the treatment with Ust. Subgroup 1 was characterized by significantly reduced ROS generation by investigated leukocyte populations along with an increase in phagocytic activity of monocytes and

neutrophils. Subgroup 2 – by moderately increased ROS production and slightly decreased phagocyte endocytosis. **Conclusion:** Treatment with Ust can causes a decrease production of ROS by circulating leukocytes in patients with PN. A high level of individual variability of the studied indices necessitates to extend the study population in a follow-up study.

Rudenko A., Mitchenko N., Kornilina E., Bavina E.

THE STATE OF LOCAL IMMUNITY INDICES OF THE GENITAL AND URINARY TRACTS IN WOMEN WITH ACUTE UNCOMPLICATED PYELONEPHRITIS

State Institution "Institute of Urology NAMS of Ukraine", Kyiv, Ukraine

miclabor@gmail.com

The infection of the genitals in women with acute uncomplicated pyelonephritis (AUP) is the leading source of urinary tract infection. To develop new approaches to the treatment of AUP pts the information on the violation of mucosal immunity of the urogenital tract in women with AUP is actual.

The aim. To study the local immunity (LI) indices of urinary and genital tracts in the presence of different pathogens.

Materials and methods. 246 AUP pts were divided into 4 groups depending on the presence of bacteria (1), associations of bacteria and mollicutes (2), mollicutes (3) or absence of pathogens (4). The control group consisted of 23 healthy women. Humoral factors were analyzed in urine and vaginal washings; phagocytic and bactericidal activity of neutrophils (Nph) and monocytes (Mc) - in scrubs from the mucous of the urethra and cervical canal.

Results. In AUP pts, a high infection of urinary and genital (67.5%, 69.5%) tracts with mollicutes (mainly *Ureaplasma* spp. – 63.8%, 62.6%) was detected. A significant increase of IgM, IgA, IgG, sIgA, SC, lactoferrin, myeloperoxidase (MPO), lysozyme (Ls), C3 and TNF- α levels from the control were established in urine, in a greater degree - in 3 group pts; in washings - levels of Ls, MPO, IgA, IgG, in 3 group pts - levels of sIgA, IgA, Ls, TNF- α were greatest compared with other groups pts. The decrease of phagocytosis and the activation of oxygen-dependent metabolism of Nph from mucosa urethra and the cervical canal in all groups pts compared to control have been noted; Mc were characterized by increased phagocytosis, decreased their absorption capacity and increased NBT-activity, but to a lesser extent than Nph, the reserve capacities of fagocytes were reduced in pts of 1-4 groups. Changes in the functional activity of Nph and Mc had the same direction in the urinary and genital tracts in AUP pts in the presence of the same spectrum of pathogens.

Conclusions. The greatest intensity of the local immunological response to inflammation was found in the analysis of humoral factors in the urine and, to a greater extent, in pts infected with mollicutes, which can be considered one of the causative factor in the recurrence of AUP.

**Rudyk M., Opeida I., Svyatetska V., Prysiashniuk A., Dovbynychuk T.,
Khranovska N., Tolstanova G., Skivka L., Shuliak A.**

**PHAGOCYTE METABOLIC PROFILE IN RATS WITH MPTP-INDUCED
PARKINSON'S DISEASE AND CONCOMITANT ULCERATIVE COLITIS**

Taras Shevchenko National University of Kyiv, Ukraine

alena_shuliak97@ukr.net

Inflammatory processes, such as chronic inflammatory bowel diseases, could be involved in the development of neurodegenerative diseases. Activation of microglial cells plays a crucial role in the pathobiology of Parkinson's disease (PD) and it is closely related with peripheral phagocyte functional state. The aim of this study was to investigate the functional state of phagocytes from different locations in rats with PD and concomitant ulcerative colitis (UC).

Wistar rats were injected subcutaneously with 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) at the dose of 20 mg/kg to induce PD-like dopaminergic neurons loss. Experimental colitis was induced by intracolonic administration of 0.1 ml of 6% of iodoacetamide on the 7th day after MPTP injections. Metabolic profile of microglial cells, circulation phagocytes and peritoneal macrophages were characterized by reactive oxygen species (ROS) generation and phagocytosis activity that were evaluated by flow cytometry. CD69 and CD14 expression by these cells was also determined. NO production and arginase activity of the cells were measured by colorimetric assay.

PD development was associated with the increase of phagocytosis intensity in microglia. The number of CD14^{high} and CD 69+ microglial cells in these animals was also dramatically increased. Peritoneal macrophages were characterised by a 3-fold increase of NO production. Microglia from rats with PD and concomitant UC were characterized by 4 time increased CD69 expression along with increased ROS and NO production. Peripheral blood monocytes of those animals upregulated CD69, while the number of CD14+ granulocytes was decreased. Decreased phagocytosis activity of circulating phagocytes was accompanied by their slightly enhanced ROS production. Peritoneal macrophages exhibited decreased NO production and enhanced arginase activity, compared to the group of animals with PD and UC.

Peripheral phagocyte metabolic activation, which was associated with UC, potentiated inflammatory process in CNS of rat with PD that was characterised by their microglial cells strong activation.

**Rudyk M.¹, Opeida I.¹, Svyatetska V.¹, Prysiashniuk A.¹, Dovbynychuk T.¹,
Khranovska N.², Tolstanova G.¹, Skivka L.¹**

**COMPARATIVE INVESTIGATION OF PHAGOCYTE FUNCTIONAL
POLARIZATION IN DIFFERENT MODELS OF PARKINSON'S DISEASE IN
RATS**

¹Taras Shevchenko National University of Kyiv, Ukraine; ²National Cancer Institute, Kyiv, Ukraine

rosiente@gmail.com

Phagocytes of innate immunity, especially microglia, are deeply involved in the pathogenesis of Parkinson's disease (PD). The local inflammation in CNS initiated by microglia is associated with phenotypic and functional changes of peripheral phagocytes. The aim of this study was to compare the functional state of phagocytes from different locations in rats with MPTP-induced and 6-OHDA-induced PD.

PD-like dopaminergic neurons loss in Wistar rats was induced with subcutaneous injection of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) or unilateral stereotaxic injection of 6-Hydroxydopamine (6-OHDA) into the striatum. Metabolic profile of microglial cells, circulation phagocytes and peritoneal macrophages were characterized by reactive oxygen species (ROS) generation and phagocytosis activity that were evaluated by flow cytometry. CD14, CD69, CD80/86 and CD206 expression by these cells was also determined. NO production and arginase activity of the cells were examined in colorimetric assay.

MPTP-induced PD in rats was associated with the large increase in CD14^{high} and CD 69⁺ microglial cells and their enhanced phagocytosis intensity. Circulation phagocytes upregulated CD69 expression, while their ROS production and phagocytosis was decreased. Peritoneal macrophages were characterised by increased NO production, CD14 and CD69 expression. In rats with 6-OHDA-induced PD, microglia showed more severe inflammatory metabolic state. Sharply decreased phagocytosis activity along with increased ROS and NO production by microglia were detected. Decreased expressions of CD14 and CD80/86, along with high CD206 expression were observed in these cells. Circulating phagocytes in rats with 6-OHDA-induced PD showed pro-

inflammatory activation. Progressive neuronal damage led to monocytosis and proinflammatory activation of peritoneal macrophages.

Inflammatory process in CNS of rat with PD was characterised by their microglia strong activation, which was more pronounced in case of 6-OHDA-induced PD. Peripheral phagocyte metabolic changes in rats with PD have different patterns of activation and could be regarded as perspective markers for early diagnostics of PD.

Senyshyn N.Yu.¹, Bychkova S.A.²

**IMMUNE AND CYTOKINE STATUS OF PATIENTS WITH ARTERIAL
HYPERTENSION AND *HUMAN PAPILOMA* VIRUS INFECTION**

¹Ivano-Frankivsk state medical university;

²Ukrainian medical military academy

svetlana_bichkova@yahoo.com

The purpose of this study was to investigate the levels of total and local immunity indexes and cytokine status in patients with arterial hypertension (AH), combined with non-alcoholic fatty liver disease (NAFLD) and *human papillomavirus* infection (HPV). The main group consisted of 76 patients with AH, combined with NAFLD and HPV in age from 35 to 55 years (mean age of 43.9 ± 5.6 years), male. The comparison group consisted of 43 patients with AH and NAFLD without any signs of HPV. Groups were randomized by age and sex. The control group consisted of 35 healthy people. It is shown that in the main group of patients have significantly higher levels of proinflammatory cytokines and transforming growth factor- β , as well as pathogenic circulating immune complexes of small size and middle size compared with patients in the comparison group, which indicates a high activity of the processes of immune inflammation. In addition, it was determined the high concentration of proinflammatory cytokines and low levels of IL-4 in saliva of patients of the main group.

Sergeeva V.S.¹, Olenov D.G.², Sergeeva L.A.², Glebova E.I.², Valchenko A.I.²

**INFLUENCE OF γ -INTERFERON AND CORTICOSTERONE ON ACTIVITY OF
SOME ENZYMES IN LEUCOCYTES OF EXPERIMENTAL RATS**

¹Doctor's office №2, Irpen, Ukraine;

²State University of Telecommunications, Kyiv, Ukraine

19sergeeva.ljubov5383@gmail.com

The experiments were carried out on 50 white non-breeding males rats weighing 180-200 g. Corticosterone was injected at doses of 2.5 and 10 mg / 100

g, γ -interferon in doses of 0.25 and 1.0 ml / 100 g of the body weight of the animal. The activity of desoxyribonuclease (DNA-ase), glycogenphosphorylase and catalase in blood's leukocytes of experimental rats was studied in comparison with intact (control animals).

The results of the experiment showed that corticosterone at a dose of 2.5 mg / 100 g did not change the enzyme activity compared to the control.

At the same time, γ -interferon at a dose of 0.25 ml / 100 g of body weight of the animal was 2.7 times, compared with the control, significantly ($P < 0.001$) increased the activity of glycogenphosphorylase; and it remained the same high after intrabdominal injection of 1.0 ml / 100 g of γ -interferon.

With increasing doses of corticosterone and γ -interferon, respectively, 2.44 ($P < 0.001$) and 1.56 ($P < 0.001$) times, the activity of DNA-ase in leukocytes increased; respectively, in 10 ($P < 0.001$) and 2.5 times ($P < 0.001$), the catalase activity in blood's leukocytes of experimental rats increased, compared with the control.

Thus, γ -interferon stimulation of cells of the immune system is accompanied mainly by activation of glycogen phosphorylase, which, when exposed on the DNA, can regulate the cell cycle, and, when exposed to the membrane, change the phenotypic features of the cell.

At the same time, immunosuppressants effects of glucocorticoids on phagocytic activity of leukocytes may be mainly by activating catalase, which reduces the formation of hydrogen peroxide.

Shilina J.V.¹, Guscha M.I.¹, Molozhava O.S.², Litvinov S.V.¹, Dmitriev O.P.¹

**ELICITATION OF *ARABIDOPSIS THALIANA* RESISTANCE AGAINST
PATHOGENIC BACTERIA BY LIPOPOLYSACCHARIDES AND SALICYLIC
ACID**

¹Institute of Cell Biology and Genetic Engineering of NAS of Ukraine, Kyiv, Ukraine;

²ESC "Institute of Biology and Medicine" of Taras Shevchenko National University of Kyiv, Kyiv, Ukraine

omolozhavaya@ukr.net

The immunocorrection method with the application of elicitors and activators of plant signaling systems is new and promising for use in agricultural production in order to increase the immune status and adaptive potential of crops. Treatment with biotic elicitors does not cause plant visible symptoms of damage and accumulation of toxic stress metabolites, but sensitizes the plant tissue, increases its resistance to the next damage. The effectiveness of protective properties of

elicitors can be increased by applying of systemic signaling molecules simultaneously with the elicitor.

The effects of combined pretreatment with lipopolysaccharide (elicitor) and salicylic acid (signaling molecule) on the disease resistance of wild-type *Arabidopsis thaliana* L. Col-0 plants and mutant lines – *jin1* (deficient in jasmonate signaling) and *NahG* (expresses bacterial salicylate hydroxylase transgene) were tested.

The lipopolysaccharide (LPS) had been isolated from saprophytic strain 8614 of *Pseudomonas aeruginosa*. Seed pretreatment by lipopolysaccharide and salicylic acid (SA) composition increased seedlings resistance to infection of *P. aeruginosa* pathogenic strain 9096.

Compared to wild-type seedlings, the protective effect was clearer in the *jin1* mutant, indicating the possibility of LPS+SA composition pretreatment to compensate JA-deficiency due to the activation of the SA-dependent signaling pathway. A lack of protective effect was observed when transgenic plants *NahG* had been treated with LPS+SA complex.

It is concluded that a pretreatment with composition consisting of elicitor and signaling molecule could affect plant cell regulatory mechanisms, in particular, may cause substitution of one certain signal pathway by another. In the case of treating *arabidopsis* with LPS+SA complex, the salicylate system is key player, as evidenced by the data about the absence of a protective effect on the salicylate-deficiency genotype *NahG*.

Siromolot A.A.^{1,2}, Kolybo D.V.^{1,2}

**IMMUNOBIOLOGICAL AND BIOCHEMICAL PROPERTIES OF
MYCOBACTERIUM TUBERCULOSIS ANTIGENS MPT63 AND MPT83**

¹Taras Shevchenko National University of Kyiv, Ukraine;

²Palladin Institute of Biochemistry of National Academy of Sciences of Ukraine
saa0205@ukr.net

With 1.8 million deaths worldwide, the World Health Organization listed tuberculosis among the top 10 causes of death in 2015. Increasing numbers of infections with multi- and extensively drug-resistant variants of the *Mycobacterium tuberculosis complex*, resistant even to newly discovered and last resort antibiotics, highlight the urgent need for understand of immunobiological properties and molecular mechanisms of action of bacteria antigens for development of an efficient vaccines and diagnostic.

In this study we focused on some of the functional properties of immunogenic antigens MPT63 and MPT83.

By immunochemistry methods was showed that *M. tuberculosis* protein MPT83 binds to the surface of U2149 macrophage-like cell line and

MPT63 specifically binds to a small subpopulation of cells within the monocyte-like line U937 (3% of the total population).

It was established that MPT63 and MPT83 can increase the percentage of cells expressing markers of macrophage activation CD11b and F4/80.

The absence of cytotoxic effects of MPT63 and MPT83 (up to 10 μ M) on cells of monocytic origin U937 and epithelial cells of A431 cell line was shown; an insignificant proliferative effect was observed on the mouse fibroblasts and cytotoxic/cytostatic effect with macrophage cells J774 after treatment with MPT63 and MPT83 was demonstrated by MTT assay. In favor of these facts long-term incubation of mycobacterial antigens with cells of epithelial and connective tissue origin did not cause obvious signs of apoptotic cell death, instead with mouse macrophages J774 led to labeling with annexin-V-eGFP.

Also we were shown non-availability changes in the concentration of U937 cells cytosolic Ca^{2+} after MPT63 and MPT83 treatment.

Thus, immunodominant proteins MPT63 and MPT83 that are synthesized in abundance by *M. bovis* or *M. tuberculosis* strains could be involved in development of tuberculosis pathogenesis.

Sokolenko V.L., Sokolenko S.V.

THE PARAMETERS OF LIPID PROFILE AND IMMUNE SYSTEM AMONG THE INHABITANTS OF THE TERRITORIES CONTAMINATED WITH RADIONUCLIDES

Bohdan Khmelnytsky National University of Cherkasy, Ukraine

sokolenko@ukr.net

We have analyzed the parameters of lipid profile and immune system in 100 students from National University of Cherkasy, who came to study from the territories of enhanced radio-ecological control. Among them there were found 50 people with the signs of vegetative-vascular dystonia (VSD) and 50 people without them. Winter examination session played the role of the additional psycho-emotional factor for students.

It was found that in the group of examined with the signs of VSD syndrome, the parameters of triglycerides and cholesterol of low density lipoproteins was higher compared to the group without VSD signs. Under the conditions of

additional psycho-emotional stress, the level of low density lipoproteins significantly increased and the level of high density lipoprotein decreased in the group of examined. The analysis of the immune system parameters showed more evident imbalance of T-cell immunity and inhibition of the phagocytic activity of monocytes in the groups with VSD signs. Correlation analysis revealed a significant interdependency between the immune system parameters and low density lipoproteins level. Particularly high coefficient values are gained in the group of examined with VSD signs in conditions of psycho-emotional stress. Growth of relative number of stab neutrophils in conditions of stress is accompanied by the increase of their correlation coefficient with the level of LDL-c. At the same time, in the group with VSD signs the phagocytic index of neutrophils decreases, which is not marked for a group without VSD signs.

In conditions of emotional stress there is a significant difference in the value of the immunoregulatory index CD4+/CD8+ between the groups with and without VSD signs, which indicates the imbalance in the activity of T-cellular immunity.

Thus, long-term living in the areas contaminated with radionuclides creates the risk of lipid metabolism dysfunctions and it can affect the parameters of immune system. Negative tendencies are intensified in conditions of emotional stress.

Stepaniuk K.S.¹, Dons'koi B.V.², Chernychov V.P.², Sudoma I.O.³

**CLINICAL AND SEASONAL ASPECTS OF NK CELL CYTOTOXICITY: 12
YEARS' EXPERIENCE IN ROUTINE TESTING**

¹Taras Shevchenko National University of Kyiv, Educational and Scientific Center "Institute of Biology and Medicine", Kyiv, Ukraine;

²Institute of Pediatrics, Obstetrics and Gynecology, National Academy of Medical Sciences of Ukraine, Kyiv, Ukraine;

³Reproductive Medicine Clinic "Nadiya", Kyiv, Ukraine

katerina.stepanyuk@gmail.com

Natural killer cell cytotoxicity (NKc) is unfavorable factor for embryo implantation and placentation. Significance and necessity of NKc testing in reproductive failures patients are still on debate.

Methods: more than 14,000 NKc test results made during 2006-2017 years were retrospectively analyzed for clinical anamnesis and age. The database of The Main Department of Statistics in Kyiv for frequency of implantation and the database of clinic "Nadia" for implantation rate (statistical databases) were investigated.

Results: Any association of NKc and frequency of elevated NKc levels with age in patients were not found. Elevated NKc was similarly founded in patients with primary(21.4%), secondary infertility(20.7%), with only 1 incident of reproductive failure(20.7%) or with anembryonic pregnancy in anamnesis(21.6%)

Increased NKc was more often detected in patients with multiple reproductive failures (RF) (2 and more IVF/PRf) – 41.1% and in patients with ectopic pregnancy in anamnesis – 34.4%.

5-8% of population demonstrated seasonal changing. The number of patients with elevated NKc was significantly decreased in autumn (26.2%) compared to spring (34.2%) and to average of the year (31.6%) during 2011-2017. Any parameters of NKc-test (target, viability, spontaneous level, effectors, quality, quantity) didn't show any seasonal dynamic.

The levels of implantation in IVF cycle and of natural implantation were significantly increased in autumn (26.4%) compared to spring (23.7%) and to average of the year (24.5%).

Conclusion: NKc had seasonal variations: decrease of NKc was associated with increase of effectiveness of implantation in IVF and implantation frequency in natural conception. No difference in NK level was found in patients with primary/secondary infertility as well as in patients with only 1 incident of RF. In contrast, the patients with multiple RF had elevated frequency of increased NKc. Elevated NKc associated with ectopic pregnancy but not with anembryonic pregnancy in anamnesis.

Stepura K.O., Opeida I.V., Rudyk M.P., Skivka L.M., Svyatetska V.M., Shulyak A.A.

METABOLIC STATE OF PERITONEAL MACROPHAGES IN RATS WITH C6 GLIOMA

Taras Shevchenko National University Educational and Scientific Centre «Institute of Biology and Medicine», Kyiv, Ukraine

cherrypie1408@gmail.com

Peritoneal macrophages play a great role in the system of mononuclear phagocytes. In the conditions of a systemic inflammatory process, these cells acquire a pro-inflammatory metabolic profile. The development of most tumor processes is characterized by general immunosuppression of the body. In conditions of glioma growth, the production of superoxide anion, hydrogen peroxide and nitrogen oxide by macrophages decreases. Therefore, a decrease in the metabolic activity of these cells adversely affects the overall

immunoreactivity of the organism. The influence of the tumor process on this line of cellular immunity can have ambiguous consequences for the organism.

The aim of the study was to investigate the metabolic state of peritoneal macrophages in rats with C6 glioma.

Wistar male rats (180-250 g, n = 20) were used in the study. C6 cells were provided by the Bank of Cell Cultures and Transplantable Experimental Tumors of R.E. Kavetsky Institute of Experimental Pathology, Oncology and Radiobiology NASU (Kyiv, Ukraine). Tumor cells were implanted intracranially into rat brain using specialized cannula that was developed in our laboratory. Metabolic state of peritoneal macrophages was determined based on NBT test, nitrite level was assayed by the Griess reaction, arginase activity was measured by colorimetric method.

Arginase activity by peritoneal macrophages in rats with C6 glioma was no change compared to intact animals. The level of nitrite production by peritoneal macrophages in tumor-bearing rat was almost halved in comparison with the control group. Spontaneous production of superoxide anion by peritoneal macrophages was suppressed in rats with C6 glioma, and production of reactive oxygen species decreased in response to stimulation of zymosan.

Thus, the obtained data testify to oppression of the functional reserve of peritoneal macrophages of tumor-bearing rats, due to changes in the metabolic state of these cells to the immunosuppressive metabolic profile under the influence of glioma.

Tymoshok N.O., Bubnov R. V. , Nechypurenko O.O. , Spivak M. Ya
**GENDER SUSCEPTIBILITY TO DICLOFENAC-INDUCED HEPATORENAL
DYSFUNCTION IN RATS**

D.K. Zabolotny Institute of Microbiology and Virology of the National Academy of Sciences of Ukraine

N_Tymoshok@ukr.net

A method of modeling Drug-induced liver injury (DILI) under diclofenac sodium (DCF) treatment rats gives the opportunity to explore the sex difference in waitlist outcomes of liver and kidney failure. In rats, inactivation of DCF occurs with the participation first phase metabolism via cytochrome P450 (Cyp3a4, Cyp2c11). However, only the masculine profile of growth hormone (GH) secretion influence on expression Cyp2c11. Mature Wistar rats 200-220 g (60 females and 60 males) were injected intraperitoneally DCF at doses of 5-10 mg / kg, females - daily for 4 days, male - twice, daily. Performed in vivo animal Doppler (ultrasound) and

determined the biochemical activity of serum alanine aminotransferase (ALT) at the beginning and the dynamics of the disease. In rat models the hepatorenal toxic effect of DCF appears to be dose-dependent and consistent with increased production of ALT in circulation and accumulation of triglycerides in hepatocytes. According to data of ultrasound at 6 weeks revealed increasing Liver chogenicity, presence of portal hypertension, expansion portal vein diameter (PVD), splenomegaly, signs of nephropathy. The following morphological patterns were observed in diclofenac induced hepatitis: the granular dystrophy of hepatocytes, lobular inflammation, sinusoid capillary stasis, hepatic sinusoids dilated especially pronounced at 12 weeks after the last injection DCF. Installed renal dysfunction, with a predominance of male sensitivity to nephrotoxic action of DCF was found, espisially atrophy of tubular epithelium, lymphocytic infiltration in renal parenchyma . The manifestations of nonalcoholic steatohepatitis extracellular and intracellular accumulation of fat have been noted to be only in females. At the same time, males (dose of 10 mg/kg) were accompanied by signs of the disease: granular dystrophy of hepatocytes, discomplexion hepatic beams, local hemorrhages, swelling of liver sinusoidal endothelial cells.

Tymoshok N.O., Bubnov R.V., Nechypurenko O.O., Spivak M.Ya.

**SEX DIFFERENCES IN LIVER AND KIDNEY FAILURE DEVELOPMENT
UNDER DICLOFENAC-INDUCED DILI MODEL IN RATS**

Zabolotny Institute of Microbiology and Virology of the National Academy of Sciences of Ukraine, Kyiv

N_Tymoshok@ukr.net

Modeling of drug-induced liver injury (DILI) under diclofenac sodium (DCF) treatment in rats gives the opportunity to study sex difference in liver and kidney failure development. DCF inactivation in rats involve activation of P450 (Cyp3a4, Cyp2c11). Masculine profile of growth hormone (GH) secretion influence on expression Cyp2c11. The aim was to study sex differences in liver and kidney failure development under diclofenac sodium (DCF) treatment in rats.

Methods: Mature Wistar rats 200-220 g (60 females and 60 males) were injected intraperitoneally DCF (5-10 mg/kg), females - daily for 4 days, male - twice, daily. Performed in vivo animal preclinical ultrasound and determined the biochemical profiles including serum alanine aminotransferase (ALT) at the beginning and the dynamics of the disease.

Results: In rat models the hepatorenal toxic effect of DCF appears to be dose-dependent and consistent with increased production of ALT in circulation and accumulation of triglycerides in hepatocytes.

The manifestations of nonalcoholic steatohepatitis extracellular and intracellular accumulation of fat have been noted to be only in females. The disease males (dose of 10 mg/kg) manifested by: granular dystrophy of hepatocytes, discomplexion hepatic beams, local hemorrhages, swelling of liver sinusoidal endothelial cells. The following morphological patterns were observed in diclofenac induced hepatitis: the granular dystrophy of hepatocytes, lobular inflammation, sinusoid capillary stasis, hepatic sinusoids dilated especially pronounced at 12 weeks after the last injection DCF.

According to ultrasound data at 6th week we revealed increasing liver chogenicity, presence of portal hypertension, expansion portal vein diameter (PVD), splenomegaly, and signs of nephropathy. Renal dysfunction was detected, with a predominance of male sensitivity to nephrotoxic action of DCF was found, espisally atrophy of tubular epithelium, lymphocytic infiltration in renal parenchyma .

Conclusions: Modeling of drug-induced liver injury (DILI) under diclofenac sodium (DCF) treatment demonstrate different sex-dependent patterns in rats in liver and kidney failure development.

Vertyporokh L.S., Hulas-Stasiak M., Kordaczuk J., Wydrych J., Wojda I.

VIRULENCE OF THE ENTOMOPATHOGENIC FUNGUS *BEAUVERIA BASSIANA* AND THE HUMAN OPPORTUNISTIC PATHOGEN *CANDIDA ALBICANS* AGAINST THE INSECT MODEL ORGANISM *GALLERIA MELLONELLA*

Maria Curie-Sklodowska University, Lublin, Poland

lydia.vertyporokh@gmail.com

The greater wax moth, *Galleria mellonella* is widely used as a model organism in the studies of innate immunity, host-pathogen interactions and human pathogens' virulence factors. The short generation time, low-cost breeding and the absence of ethical restrictions encourage scientists to replace small mammals with insects in the immunological studies. It is possible due to similarities and the common origin of the immune system of vertebrate and invertebrate animals. The entomopathogenic fungus *Beauveria bassiana* is a natural pathogen of *G. mellonella*, so these two organisms are linked by the long history of coevolution. Therefore, during *B. bassiana* infection we may anticipate the activation of the

specific immune response of *G. mellonella*, while the opportunistic pathogenic fungus *Candida albicans* likely initiates unspecific general immune mechanisms. We aimed to compare the infection process in *G. mellonella* larvae caused by *B. bassiana* and *C. albicans* fungi.

We studied the *G. mellonella* humoral immune response (phenoloxidase activity, antifungal activity of hemolymph, defense peptides expression) and histological specimens after the infection with *B. bassiana* and *C. albicans*. We revealed numerous differences between the infection process caused by the natural and opportunistic pathogens. The infection with *B. bassiana* had higher efficiency in the overcoming of the host defense than *C. albicans* infection: we observed destruction of the fat body, an organ responsible for the defense peptides synthesis, accompanied by the dose-dependent changes in the defense peptides expression.

We believe that when focusing only on natural or only on non-natural pathogens we may overlook some important properties of the immune response of the studied host. The infection with natural and opportunistic pathogens provide to different outcomes that is probably linked with the ability of natural pathogens to escape recognition and target the immune system of the host.

Vertyporokh L.S., Kordaczuk J., Hulaś-Stasiak M., Wojda I.

SPECIFICITY OF *GALLERIA MELLONELLA* IMMUNE PRIMING WITH HUMAN OPPORTUNISTIC PATHOGEN *CANDIDA ALBICANS*

Maria Curie-Skłodowska University, Lublin, Poland

lydia.vertyporokh@gmail.com

The insect immune system shares many features with the innate immune system of vertebrates, but insects lack adaptive immunity based on antibody-antigen pairs. However, since the 1990s scientists have been reporting the kind of “immune memory” in insects. This phenomenon is called immune priming and its mechanisms remain unclear. One observes immune priming when the previous contact with the pathogen results in enhanced resistance to the same (homologous priming) or other (heterologous priming) pathogen in the future. Immune priming attracts great interest and there are attempts to find the similar phenomenon within human immune system, so-called trained immunity.

The greater wax moth, *Galleria mellonella*, is a popular model in immunological studies with well-described immune system. We aimed to investigate the specificity of immune priming in *G. mellonella* larvae within one generation. We used various natural and non-natural fungus and bacterial

pathogens of *G. mellonella*. After injection of the non-lethal dose of the pathogen the larvae were infected with the lethal dose of the same or other pathogen, and survival rates were measured. So far, we observed only homologous immune priming for the one of the tested pathogens – *Candida albicans*. We present some aspects of host-pathogen interaction in the primed and non-primed animals. The further studies are needed to elucidate the mechanisms of the activation of such specific response.

Voloshchuk O.N., Kopylchuk G.P.

**CELLULAR IMMUNITY STATE UNDER TOXIC HEPATITIS ON THE
BACKGROUND OF ALIMENTARY PROTEIN DEFICIENCY**

Yuriy Fedkovych Chernivtsi National University, Chernivtsi, Ukraine

o.voloshchuk@chnu.edu.ua

Studies on the role of immunity mechanisms in the emergence and maintenance of inflammatory and destructive processes in the liver under toxic hepatitis and nutrient deficiency are topical.

The aim of research – to study the quantitative content and functional activity of leukocytes under the conditions of acetaminophen-induced hepatitis on the background of nutritional protein deficiency. The animals were separated into the following experimental groups: I – animals receiving complete semi-synthetic ration (C); II – animals receiving low-protein ration (LPR); III – animals subjected to acetaminophen-induced liver lesions receiving complete ration (H); IV – animals subjected to acetaminophen-induced liver lesions that were previously fed semi-synthetic low-protein ration (LPR+H).

The most pronounced changes in cell-mediated immunity are observed in protein-deficient animals with toxic hepatitis. The pronounced defects of both specific and non-specific cellular immunity were manifested by the leukocytosis, increase number of segmented neutrophils in blood serum against decrease their phagocytic index and phagocytic number, reduction of total lymphocyte number, and simultaneously lowering of T- and B-lymphocytes was established under the conditions of acetaminophen-induced hepatotoxicity on the background of protein deficiency. Installed changes indicate the defective formation of functional immunity state which can manifest by decrease the body's ability to carry out the reaction of cellular and humoral immunity.

Research results may be used for the rationale of therapeutic approaches to the elimination and correction of the consequences of immunological status disturbances under the conditions of acetaminophen-induced hepatitis, aggravated by the alimentary protein deprivation.

**Yakovenko L.F.¹, Tsisarenko A.M.¹, Vikarchuk M.V.², Grygorenko V.M.²,
Pogribnyy P.V., Kroupskaya I.V.¹**

**ANTIBODIES AGAINST HEAT SHOCK PROTEIN 60 IN PROSTATE CANCER
PATIENTS**

¹Institute of Molecular Biology and Genetics of National Academy of Sciences of Ukraine, Kyiv, Ukraine;

²State Institution "Institute of Urology of National Academy of Medical Sciences of Ukraine", Kyiv, Ukraine

I.f.yakovenko@imbg.org.ua

The detection of anti-Hsp60 antibodies may have clinical usefulness in screening, diagnosis and prognosis of cancer. The role of anti-Hsp60 antibodies at malignant diseases is largely unknown. The aim of this work was to study the influence of IgG antibodies affinity purified from serum samples of prostate cancer patients and blood donors on the viability of LNCaP cells in vitro.

Materials and methods. Fifty five patients with localized, advanced and generalized prostate cancer who had undergone radical prostatectomy were selected. Serum reactivity to Hsp60 in patients was determined by ELISA and Western blotting. Obtained and purified recombinant prokaryotic (GroEL) and human Hsp60 were used as antigens. Donor's serum samples with low reactivity to Hsp60 were used as control. LNCaP cells were treated with IgG antibodies affinity purified from serum samples of patients with aggressive disease and blood donors in different concentrations. Dehydrogenases activity and changes in signaling pathways of LNCaP cells were measured by MTT Assay and Western blotting respectively.

Results. There was significant difference in the serum levels of IgG anti-GroEL antibodies between patients and control by ELISA ($p < 0.05$). High serum reactivity to GroEL or human Hsp60 was detected in all patients (11/11) with a biochemical recurrence after radical prostatectomy by Western blotting. Among such patients, 36.36% (4/11) had a PSA level below 10 ng/ml, and 45.45% (5/11) had Gleason score below 7 before surgery. High serum reactivity to human Hsp60 was observed at locally advanced and generalized cancer in patients who had not a biochemical recurrence during the follow-up period. IgG anti-Hsp60 antibodies purified from serum samples of patients and blood donors influenced on dehydrogenases activity of LNCaP cells, heat shock proteins and Erk 1/2, Akt1, p70S6K kinases expression in different way.

Conclusion. IgG anti-Hsp60 antibodies affinity purified from prostate cancer patients with aggressive disease were found to influence on the viability of LNCaP cells in vitro.

Yanovska V.G., Tryliska T.V., Trachuk Z.G.

**ANALYSIS OF THE DETECTION OF MARKERS FOR SYSTEMIC
AUTOIMMUNE DISEASES IN PEDIATRIC PRACTICE**

¹Ukrainian Reference Centre for Clinical Laboratory Diagnostics and Metrology of OKHMATDYT National Children's Specialized Hospital of Ministry of Health (MoH) of Ukraine, Kyiv, Ukraine

trilltatiana@gmail.com

BACKGROUND: Autoimmune diseases (AD) are characterized by abnormal response of body's immune system against its own healthy tissues. As a result, own body cells are perceived as foreign antigens, and specific antibodies (Ab) are produced for their neutralization and elimination. AD are divided into two groups: organ-specific (OSAD) and systemic (SAD). The main hallmark of AD is presence of auto-antibodies with different antigenic specificity. Auto-antibody detection is used to diagnose and differentiate OSAD and SAD. **METHODS:** The results of laboratory examination of pediatric patients with SAD who were on the treatment in OKHMATDYT National Children's Specialized Hospital of Ministry of Health (MoH) of Ukraine are presented. IgG with different specificity (nRNP/Sm, SS-A, Ro-52, SS-B, Scl-70, Jo-1, PCNA, PM-Scl, ribosomal P proteins, centromeres, native dsDNA, cyclic citrullinated peptides) were used as a markers of SAD and were examined by enzyme immunoassay using automated enzyme immunoassay analyzer. **RESULTS:** According to the diagnosis, pediatric patients were divided into four groups: systemic lupus erythematosus, n=17, age range 5-17 years; dermatopolymyositis, n=9, 3.5-16 years; juvenile rheumatoid arthritis (JRA), n=146, 1-18 years; diffuse connective tissue diseases (DCTD), n=22, 1.7-15 years. IgG auto-antibodies were revealed only in 21 pediatric patients with different AD: SLE – 35,29% (n=6), age range 12-17 years; JRA – 8,22% (n=12), 3.5-16 years; DCTD – 13,64% (n=3), 5-16 years. There were no positive results of auto-antibody detection in patients with dermatopolymyositis, although the list of investigated antinuclear Ab included anti-Jo-1 IgG, that is specific marker for this pathology.

CONCLUSION: Very low detection rate of IgG auto-antibody in younger pediatric patients with SAD was revealed. In patients with SLE auto-antibodies can be detected from the age of 12 years, with JRA and DCTD - from 3.5 years.

We suppose that age-related immune system immaturity is the main reason of this phenomenon. Therefore, clinical symptoms and the results of complex laboratory investigation remain the main criteria in the diagnostics of SAD in pediatric patients.

Zagorodnya S.D.¹, Baranova G.V.¹, Maksymenok O.V.²

**STUDY OF PATTERNS OF IMMUNE RESPONSE TO HIV-ASSOCIATED
EPSTEIN-BARR VIRUS IN PATIENTS WITH AIDS**

¹D.K. Zabolotny Institute of Microbiology and Virology, NASU, Kyiv Ukraine;

²SI "Lev Gromashevski Institute of Epidemiology and Infection Diseases, NAMSU, Kyiv Ukraine

svetazagorodnya@ukr.net

Studies have been conducted on the content of specific antibodies to the Epstein-Barr virus (capsid and nucleic antigens) in blood serum of patients with HIV in different groups, namely, HIV-infected people who have no illness and manifestations of AIDS, HIV-infected injected addicts, HIV infected children. An analysis of the correlation of antibodies of IgG or IgM classes with the help of the "IFA-AtVEB-strip" test system, developed at the Institute of Microbiology and Virology of the National Academy of Sciences of Ukraine, and comparison with the reference data of test systems with differentiation of immunogenic antibodies to the EBV (capsid and nucleic antigen) was performed.

As a result of the performed studies, it was found that HIV-infected injected addicts and HIV-infected pregnant women have the highest percentage of specific antibodies to the Epstein-Barr virus, that is, a clear reactivation of the Epstein-Barr virus against the background of immunosuppression of the organism, which is exacerbated by drug use and the women body condition during pregnancy. It should be noted that only 1,5% of the tested pregnant women who were not infected with HIV had a reactivation of EBV infection, and in HIV-infected people who did not use drugs, the level of antibodies to EBV was 60% lower.

The surveyed group of children with HIV infection had an increased level of antibodies to the capsid antigen of the Epstein-Barr virus (70% vs. 50%) and reduced the level of nucleic antibodies (58% vs. 70%).

The obtained data allowed to reveal new data on the interaction of human immunodeficiency virus and virus Epstein-Barr in blood serum of HIV-infected children and sick HIV-infected injected addicts.

Zantaraia T.¹, Popyk A.², Tsymbaliuk R.¹, Opalchuk K.¹, Susak Y.¹
SERUM LEVEL OF HIGH MOBILITY GROUP BOX 1 PROTEINS IN PATIENTS
WITH SEVERE ACUTE PANCREATITIS IN THE COURSE OF TREATMENT
WITH ULINASTATIN

¹Bogomolet's National Medical University, Kyiv, Ukraine;

²Taras Shevchenko National University, Kyiv, Ukraine

zxcv392@ukr.net

Background: Severe acute pancreatitis (SAP) sets in as a local inflammation of pancreatic tissue, and progresses with the development of pancreatic necrosis, systemic inflammation and multiple extrapancreatic organs dysfunction. Ulinastatin (Ust) is reported to successfully use in the complex therapy of SAP. USt is a serine protease inhibitor with anti-inflammatory properties. In experimental models of SAP and other inflammatory diseases, USt exerts anti-inflammatory effect by increasing the proportion of Tregs as well as by downregulating High Mobility Group Box 1 protein (HMGB1) expression. A positive association between serum HMGB1 levels and the progression of pancreatitis has been reported. HMGB1 inhibitors represent a promising agents for the treatment of inflammatory diseases including SAP. The aim of this work was to evaluate serum level of HMGB1 in patients with SAP in the course of treatment with USt. **Methods:** Serum level of HMGB1 was measured by the commercially available HMGB1 ELISA Kit (Elabscience, USA). **Results:** All patients with pancreatitis had higher serum level of HMGB1 than that in healthy persons. For obtained data analysis, we have used literature data concerning serum concentration of the alarmins in healthy volunteers, since we had not the group of healthy persons in our own experiments. Treatment of patients with SAP causes the decrease of HMGB1 serum level. However, the difference in pre and post treatment data was only statistically significant in patients whose treatment included ulinastatin. **Conclusion:** Significant individual variability of serum HMGB1 level in SAP patients before and after the treatment suggests the possibility to use this criterium as a marker for the personalized disease course monitoring and for individualized evaluation of treatment efficacy. The evaluation of serum HMGB1 levels need to be supplemented with the examination of additional inflammation markers such as cytokines and/or cellular indices of local and/or systemic inflammation.

Zelenska A.D., Kolyada T.I.

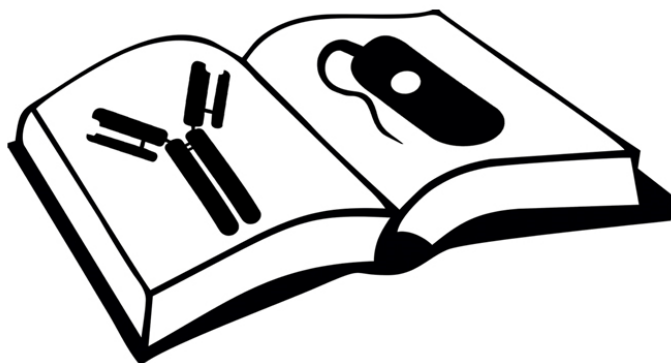
**VIRAL INFECTION AS A KEY ELEMENT IN THE PATHOGENESIS OF
MULTIPLE SCLEROSIS**

Mechnikov Institute of Microbiology and Immunology of the National academy of medical sciences of Ukraine, Kharkiv, Ukraine

labimmun@gmail.com

Multiple sclerosis (MS) is an autoimmune demyelinating disease of the central nervous system (CNS), and its development is associated with the action of a large number of pathogenetic factors. In recent years, ample evidence supports the hypothesis of the key pathogenetic role of the Epstein-Barr virus (EBV) and some retroviruses. In addition to epidemiological data, association of EBV with MS is indicated by a significant increase in IgG titres to EBV antigens, mainly to EBNA-1, in serum of patients a few years before the onset of clinical manifestations of the disease [Lünemann J.D. et al., 2010]. EBV-infected B cells were detected in white matter lesions in post mortem brain samples of patients with MS [Serafini B. et al., 2007]. Serafini B. et al. found that CD8+ T cells infiltrated all the sites in the CNS where infected B cells were located, and the number of CD8+ T cells strikingly correlated with the number of EBV-infected B cells, including active foci of demyelination in acute MS cases. Mameli G. et al. showed that binding of EBV induces the activation of human endogenous retroviruses MSRV/HERV-W in peripheral blood mononuclear cells and in astrocytes [Mameli G. et al., 2012]. In infectious mononucleosis, the increased expression of MSRV/HERV-W in peripheral blood mononuclear cells has been observed, moreover, a direct correlation has been found between levels of IgG to EBNA-1 and levels of MSRV-specific mRNA expression [Mameli G. et al., 2013]. Activation of MSRV/HERV-W was revealed in inflammatory context and in neuropathogenic processes in MS [Morandi E. et al., 2017; Mameli G. et al., 2007]. Thus, EBV infection and activation of retroviruses are considered as key elements in the pathogenesis of MS. Within the framework of the "viral hypothesis", the most important tasks are the verification of data indicating the possible etiological role of EBV, the study of the pathogenetic mechanisms associated with MSRV/HERV-W at different stages of MS development, as well as the identification of immunological and genetic factors associated with the defective control of EBV-infected B cells and, as a result, their migration and accumulation in the CNS.

SECTION



**METHODICAL
FUNDAMENTALS OF TEACHING OF
MICROBIOLOGY AND IMMUNOLOGY**

Butov D.O.¹, Kuzhko M.M.², Stepanenko H.L.¹, Butova T.S.³

ASSOCIATION BETWEEN IL-4 GENE POLYMORPHISMS AND RPOB GENE OF *MYCOBACTERIUM TUBERCULOSIS* IN PATIENTS WITH MULTI-DRUG-RESISTANT TUBERCULOSIS

¹Kharkiv National Medical University, Kharkiv, Ukraine;

²National Institute on Phthisiology&Pulmonology named by F.G. Yanovsky NAMS of Ukraine, Kiev, Ukraine;

³V. N. Karazin Kharkiv National University, Kharkiv, Ukraine

dddima@gmail.com

Background and objective. Determine associations between IL-4 gene polymorphisms and rpoB gene of *Mycobacterium tuberculosis* (MTB) in patients with multi-drug-resistant tuberculosis (MDRTB).

Methods. The study included 117 Caucasian people with pulmonary tuberculosis (TB) including 47 patients with MDRTB and rpoB gene of MTB (1stgroup), 40 without MDRTB (2ndgroup) and 30 healthy donors (3rdgroup). Serum levels of IL-4 were evaluated by ELISA. Studied promoter region C589T of the IL-4 gene. Detection of MTB complex DNA and rpoB gene was performed using XpertMTB/RIF Assay.

Results. In the 1stgroup the levels of IL-4 were (7.15 ± 0.82) , 2nd – (12.02 ± 0.69) and 3rd – (29.99 ± 1.27) pg/L ($p < 0.001$). Homozygote CC genotype of the IL-4 gene predominated in 3rdgroup was $56.67 \pm 9.05\%$ ($N=17$) compared to patients with TB: 1stgroup – $0.00 \pm 0.02\%$ ($N=0$) and 2nd – $5.00 \pm 3.45\%$ ($N=2$). Heterozygous CT genotype of the IL-4 gene was observed in $97.87 \pm 2.10\%$ ($N=46$) in 1st, $20.00 \pm 6.32\%$ ($N=8$) in 2nd and $23.33 \pm 7.72\%$ ($N=7$) in 3rdgroups. Homozygous TT genotype of the IL-4 gene was $2.13 \pm 2.10\%$ ($N=1$) in the 1stgroup, in 2nd – $75.00 \pm 6.85\%$ ($N=30$) and 3rd – $20.00 \pm 7.30\%$ ($N=6$) ($p < 0.001$).

Conclusion. Compared to healthy controls patients with TB had significantly high levels of serum IL-4. This coincided with greater frequency of heterozygous CT genotype in 1stgroup and homozygote TT genotype in 2ndgroup polymorphism C589T of the IL-4 gene. In addition in patients with MDRTB and with the presence of rpoB gene of MTB was discovered close relationship with heterozygous CT genotype of the IL-4 gene. Further studies are warranted whether higher rate of MDRTB has a causal immunogenetic relationship to polymorphism of gene encoding for IL-4 than patients without MDRTB. In addition, these studies revealed a significant influence of the polymorphism C589T gene IL-4 on the changes in the population of Th-lymphocytes, clinical

symptoms, relapse of tuberculosis, formation destructions in the lung, which may treatment outcomes in patients with MDRTB.

Demchenko N.R., Tkachenko S.V., Tretyak A.P.

THE PECULIRITIES OF MICROBIAL CORROSION OF STEEL, INDUCED WITH MONO- AND MIXED CULTURES OF SULFATE-REDUCING BACTERIA

National T.H.Shevchenko University "Chernihiv Collegium" 53, Hetmana Polubotka str., Chernihiv, 14013, Ukraine

nata_demch@ukr.net

The process of microbial steel corrosion occurs at presence of aggressive microbial community, in which sulfate-reducing bacteria (SRB) prevail. The main metabolite of SRB is hydrogen sulfide, which intensifies metal destruction. Among the components of microbial community various metabolic bonds are established, influencing the formation of corrosively active biocenosis.

The aim of this paper is to define the peculiarities of microbial corrosion of steel, induced with mono- and mixed cultures of sulfate-reducing bacteria.

The process of microbial steel corrosion was studied in mono- and mixed cultures of SRB. As monocultures, the SRB of *Desulfovibrio* sp. M-4.1 and *Desulfomicrobium* sp. TC 4. strains were used. The mixture of *Desulfomicrobium* sp. TC 4 and *Desulfovibrio* sp. M-4.1 was taken in the ratio of 50 %:50 %; 70 %:30 %; 30 %:70 % correspondingly. The research was conducted using microbiological, corrosive, chemical and analytical methods.

It has been defined that under the microbial steel corrosion, the bacteria of *Desulfovibrio* sp.M-4.1 demonstrate higher corrosive (by 1,53 times) and sulfate-reducing (by 1,45 times) activity compared to the bacteria of *Desulfomicrobium* sp. TC 4 strain.

The rates of microbial steel corrosion (by 2,0-4,6 times) and hydrogen sulfide production (by 2,6 – 4,4 times) are higher in mixed SRB cultures compared to individual strains. The corrosive activity of the mixed SRB culture of *Desulfomicrobium* sp. TC 4 and *Desulfovibrio* sp. M-4.1 with the ratio of 30%:70% is 1,5 times higher that of the cultures with different combination of strains. This is consistent with the highest quantity of adhered and plankton SRB cells in the mixed culture of *Desulfomicrobium* sp. TC 4 : *Desulfovibrio* sp. M-4.1 in the ratio 30%:70% correspondingly.

Hereby, the SRB of *Desulfovibrio* sp.M-4.1 strain have been defined as more corrosively active than those of *Desulfomicrobium* sp. TC 4. strain. The SRB of

Desulfovibrio sp.M-4.1 strain creates a more corrosive medium in the composition of corrosive microbial community.

Klonovets A.A., Lifar M. H., Tretyak A.P.

PHENOTYPE CHARACTERISTICS OF IRON-REDUCING AND DENITRIFYING BACTERIA, ISOLATED FROM THE FERROSPHERE

National T.H.Shevchenko University "Chernihiv Collegium" 53, Hetmana Polubotka str., Chernihiv, 14013, Ukraine

nata_demch@ukr.net

The biggest contribution to the steel corrosion processes, which occur in the biofilm, in the subterranean medium is made by sulfate-reducing bacteria. Denitrifying, ammonifying and iron-reducing bacteria as their heterotrophic satellites play an important role in the biofilm structure formation. Hence, it is relevant to isolate and study the microorganisms of corrosively active microbial community, which participate in the processes of metal corrosion.

The aim of this paper is to describe morphological, as well as physiological and biochemical properties of certain iron-reducing and denitrifying bacteria, isolated from the ferrosphere of corroded pipeline.

Iron-reducing and denitrifying bacteria were isolated in the Kalinenko and Hiltay media correspondingly. Multiple seeding of certain colonies onto corresponding liquid and solid nutrient media were carried out to obtain pure cultures. Pure cultures were isolated using the Koch method.

Light and phase contrast microscopy with magnifying power ($\times 1000$) and electronic microscopy.

Morphological and physiological and biochemical properties of the isolated strains were studied according to the generally accepted methods.

The pure cultures of iron-reducing (IRB) and denitrifying bacteria (DNB) were isolated from the ferrosphere, formed on the steel surface. It has been established that the cells of the IRB strain 1 are represented by separate bacilli with rounded ends. The bacteria are gram-positive, they are mesophiles and aerobes, they do not produce spores. The bacteria do not demonstrate proteolytic activity, do not produce indole, ammonia, produce hydrogen sulfide, do not produce acetylmethylcarbinol, produce ornithinedecarboxylase, lysinedecarboxylase and argininedehydrolase, assimilate glucose and sucrose.

The cells of IRB strain 4 are round-shaped, gram-positive, mesophiles, have capsules, do not produce spores, optional anaerobes. The bacteria demonstrate proteolytic activity, do not produce ammonia, hydrogen sulfide, produce indole, do

not produce acetylmethylcarbinol, produce argininedehydrolaze, do not produce ornithinedecarboxylase, lysinedecarboxylase, assimilate glucose and sucrose.

Klymnyuk S. I., Romanyuk L. B., Kravets N. Y.

THE USAGE OF PRACTICALLY ORIENTED TRAINING IN THE TEACHING OF MICROBIOLOGY, VIROLOGY AND IMMUNOLOGY AT THE DEPARTMENT OF THE I. HORBACHEVSKIY TERNOPIL STATE MEDICAL UNIVERSITY

I. Horbachevskiy Ternopil State Medical University, Ternopil, Ukraine

kravetc@i.ua

Integration of Ukraine into the European educational space on the one hand and the marked shortage of doctors in the European post-Soviet countries encourage us to reform the systems of training specialists in the medical and biological directions. According to the fact, that microbiology, virology and immunology are one of the fundamental subjects for the preparation of students of the medical faculty, due to the teaching experience of previous years; our department focuses on practically oriented training, namely the development and delivery of practical skills by students.

For successful result each section of discipline ends with the final occupation, which is the solution to the tests from the base of "Step-1", practical tasks (depending on the specificity of the section) and the passing of practical skills. Every student receives an individual task, which is one of the links of bacteriological or serological diagnostics of certain pathology, executes it practically and verbally interprets the probable data obtained.

The laboratory department's assistants for the final lessons prepare a special set of reagents and demonstration tests to provide each student in the group. The patterns of practical skills tasks have been prepared by the teaching staff, which briefly describes the main stages of its implementation.

Such approach of the acquisition of practical skills, namely, the individual execution of certain laboratory manipulations, will allow improving knowledge at the cognitive and tactile levels and acquiring cognitive, technological, informational and predictive, analytical and professional competencies. Besides, practically oriented training will allow students to feel more fluent and confident during classes in clinical departments, in particular infectious diseases, therapy, paediatrics, functional and laboratory diagnostics, primary health care and general practice of family medicine; deepen interdisciplinary connections and will enable future specialists to understand the need for integration of theoretical knowledge into practical medicine already in the first year.

Kolodii S.A., Kordon J.V., Kovalenko I.M.

THE USING OF TEST CONTROL FOR STUDY OF MICROBIOLOGY

Vinnitsya National Pirogov Memorial Medical University, Vinnitsya, Ukraine

kovalenko.in@gmail.com

The included of Ukraine in the European system of higher education accompany transformation processes. Higher school of our country has a goal to prepare competitive specialists. The pedagogical collectives of medical institutes of higher enter modern educational technologies of studies with the use of analytically-searching work and scientific information. An important task is introduction of new technologies of studies, presentation of them on a new high-quality level, embodiment of them in practice of collectives of departments.

Study of microbiology in preparation of doctors it is necessary for a fight against infections. Knowledge from microbiology are base for clinical disciplines, as assist logical perception of clinical data, form clinical thought without which it is impossible to become a highly skilled specialist.

For the improvement of quality of preparation of specialists there is a necessity of application of modern methods of studies, control, which provide the increase of creative activity of students, sent to forming and development of professional thought.

The continuous checking of knowledge of students system is widely used . To that end test tasks geared-up on the topic of every practical employment (current control of initial level of knowledge). Writing test control is conducted at the beginning of employment, occupies 7-10 minutes and allows to define the initial level of preparation of every student.

Test control provides simultaneous verification of knowledge of students of all group and forms for them motivation for preparation to every employment.

Main advantage of tests is the fully automated verification of knowledge of students, which provides maximally possible her

Current verification is this studies, with fixing, reiteration and analysis of educational material. With the purpose of exposure of end-point of studies it is necessary to apply final control on which it is possible to judge students about general achievements.

Kryzshanovskaya A. V.

**METHODICAL APPROACHES IN TEACHING MICROBIOLOGY TO STUDENTS
OF STOMATOLOGICAL FACULTY**

National Pirogov Memorial Medical University, Vinnytsya, Ukraine

2205avk1965@gmail.com

The system of teaching microbiology, virology and immunology at the Stomatological Faculty plays an important role in the formation of future specialists. At lectures, students receive information about the research outline of pathogens of oral cavity diseases, the basis of asepsis, antiseptics, sterilization, disinfection, the role of nonspecific and specific factors in the local protection. During practical classes, students consolidate the theoretical material and acquire practical skills. At the same time, our discipline is replenished with up-to-date information about new biological variants of pathogens, the invention of new pathogens and about mechanisms of antimicrobial resistance to antimicrobial drugs. This situation requires the solving of new problems in the study of the infectious process at the molecular, cellular, tissue, organ levels and their interconnections. One of the main areas of studying microbiology is the etiological structure of infectious pathology and its change in the genesis of infectious diseases. The methodical development of this direction introduces students to the mechanisms of their various formations, which they will encounter in their practice. The microbiological, immunological and virological research can establish the relationship between the pathogens of the infectious disease and the immune system of the patient's body. It has a leading role in shaping the professional outlook of future professionals. The basis of the methodology of teaching microbiology is the principle of pathogenetic, clinical and epidemiological study of pathogens. Methodically correct is to allocate microorganisms that cause similar diseases (surgical, hospital infection, respiratory diseases and intestinal infectious diseases) into a single group. From the point of view of epidemiology it is important to determine the pathways for the transmission of pathogens. Therefore, airborne, intestinal, contact and other diseases that characterize the pathways and mechanisms of transmission of their pathogens are emitted. Thus, the teaching of microbiology, virology and immunology is carried out using methodological foundations of higher education pedagogy.

Rudenko R.Y., Demchenko N.R.

THE SENSITIVITY OF SULFATE-REDUCING BACTERIA OF *DESULFOVIBRIO* SP. M 4.1 AND *DESULFOMICROBIUM* SP. TC 4 STRAINS TO QUINOLINE DERIVATIVES.

National T.H.Shevchenko University "Chernihiv Collegium" 53, Hetmana Polubotka str., Chernihiv, 14013, Ukraine

nata_demch@ukr.net

One of the possible ways to solve the problem of metal constructions protection against microbial corrosion is the search of the substances with biocidal activity against sulfate-reducing bacteria – the most aggressive component of sulfidogenic microbial community of ferrosphere. The compounds, containing tetradic Nitrogen, act as effective biocides against sulfate-reducing bacteria. Nowadays, a few substances are known to be used as biocides-inhibitors of microbial corrosion. Therefore, the search of the substances with antimicrobial properties against sulfate-reducing bacteria is relevant. The aim of this paper is to study the sensitivity of sulfate-reducing bacteria of *Desulfovibrio* sp. M.4.1 and *Desulfomicrobium* sp. TC 4 strains to tetradic quinoline salts.

The biocidal properties of tetradic quinoline salts (concentration 1%) were defined with the help of hole method in the volume of agar Postgate 'B' medium previously inoculated with the suspension of sulfate-reducing bacteria. The diameter of bacteria growth retardation zone was used to define sensitivity of the bacteria to the substance.

The results of the study have shown that the sulfate-reducing bacteria of both strains are sensitive to the impact of tetradic quinoline salts. The diameters of bacteria growth inhibition at substance were 18,0 – 32,0 mm, which confirms the high biocidal activity. The bacteria of *Desulfomicrobium* sp. TC 4 have been proven to be more sensitive to the impact of substances with methylphenacyl, brominephenacyl and etoxiphenacyl fragment. The zones of bacteria growth retardation at substance were 28,0 - 30,0 mm. The bacteria of both studied strains have been found to be equally sensitive to the impact of tetradic quinoline salts, containing nitrophenacyl, penthylphenacyl and cyclehexylphenacyl fragments. The zones of bacteria growth retardation at substance were 18,0 - 25,0 mm(. The level of substance lipophilicity has been established to have a considerable effect on the expression of the antibacterial action, namely, with the increase of the lipophilicity the antimicrobial activity decreases.

Ivaschenko N.V.**SCIENTIFIC ACTIVITIES HIGHLIGHTING OF THE UKRAINIAN SCIENTISTS IN
THE CONTEXT OF VETERINARY MEDICINE THOUGH BY
BIOBIBLIOGRAPHICAL PUBLICATIONS (2006–2013)**

State Scientific Control Institute of Biotechnology and Strains of Microorganisms
(SSCIBSM) Kyiv, Ukraine

natali-iva@ukr.net

Have been published four biobibliographical references with the research library of the State Scientific Control Institute of Biotechnology and Strains of Microorganisms and National scientific agricultural library during 2006-2013.

Under the head “Corresponding members of NAAS” – “Anatoliy Holovko” (2006), “Vyacheslav Herman” (2011), “Valeriy Ushkalov” (2013). Under the head “Academicians of NAAS” – “Anatoliy Holovko” (2011).

The scientific way of aforecited scientists is started at the Ukrainian research institution of veterinary medicine (today – National scientific center “Institute of Experimental and Clinical Veterinary Medicine”, Kharkiv) and continue at the State Research Control Institute of iotechnology and Strains of Microorganisms (Kyiv).

Under the Mr. Holovko direction were passed 8 Ph.D. defenses; 5 – PhD thesis's; published more than 400 scientific papers till 2011.

Under the Mr. Ushkalov direction were passed 7 Ph.D. defenses and published 450 scientific papers till 2013.

V. Herman's academic heritage is more than 250 scientific papers. Was prepared cohort of scientists by the academic – the whole scientific school which accounts 27 postgraduates and postgraduate students.

This way, biobibliographical references is help to researchers to find and use necessary resources of published information by means of academic paper check list. The represented information can be used as reference of scientific history, at the research and practice papers of the scientists, teachers, students and Ph.D. candidate.

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