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ABSTRACT BOOK

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SELECTION OF PRIMERS FOR HEMAGGLUTININ, NEURAMINIDASE, AND NUCLEOPROTEIN INFLUENZA A SUBUNIT GENES OF SUBTYPES H1N1 AND H7N9. PCR - RFLP METHOD OF RAPID DIAGNOSIS

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Aim. Today, influenza A viruses occupy one of the leading positions in the structure of the incidence of infectious diseases. Despite the supervision and preventive measures carried out by the World Health Organization, this disease continues to be relevant and socially significant. Given the large variability of the genome of influenza A viruses, when identifying it, it is necessary to use primers that correspond to the most conserved parts of the viral genome. The basis for the search for primer annealing sites was a comparison of the nucleotide sequences of the hemaglutinin, neuraminidase, and nucleoprotein genes of all influenza A viruses of the H1N1 and H7N9 subtypes presented in the international database GenBank (National Centre of Biotechnology Information). The **aim** was to determine [the] primers and develop an express method for the diagnosis of [the] influenza virus subtypes H1N1 and H7N9 by PCR - RFLP. **Methods.** The most conserved regions of the hemaglutinin, neuraminidase, and nucleoprotein genes H1N1 and H7N9 subtypes were selected as targets for the selection of primers. Primer design was performed based on sequence alignment of the HA, NA, and NP genes of the H1N1 and H7N9 subtypes presented in GenBank. Alignments were performed using the Lasergene (version 6.0.), BioEdit (version 7.0), and Amplify (version 1.06 Univers of Wisconsin, Genetics, Madison) programs. The melting temperatures of the primers were calculated using the Oligonucleotide Properties Calculator program (http://www.basic.northwestern.edu/biotools/OligoCalc.html). **Results.** The following pairs of oligonucleotides were obtained: HA5-ACACCAGCCTCCCATTTCAG and CCCCCTCAATAAAGCCAGCA; HA10-GCCGCAAATGCAGACACATT and GCTGCCGTCACACCTCTATT; NA1-CAGGAGCCCATATCGAACCC and CTTTGGGTCGCCCTCTGATT; NA8-TGCAGGGATAACTGGCATGG and GCTCCCGCTGTCCAGATTG;[.] NP5-GTGGTCAGCCTGATGAGACC complementary to at least 95% of the HA, NA, and NP viral RNA genes plays an important role in the development of a PCR test for rapid identification. When determined in the samples analyzed, the PCR products (RNA fragments) of 958 n.s. were obtained for the primers specific for H1N1 and of 966 n.s. for [the] HA5 primers specific for H7N9. For HA10 - 416 n.s. for H1N1 and 411 for H7N9. For NA1 (H1N1), 845 n.s. and 848 for H7N9, for NA8 (H1N1) 450 n.s. and 447 for H7N9. **Conclusions.** The lack of homology with other viruses of the Orthomyxoviridae family and with other subtypes of influenza A virus was the main criterion for the selection of oligonucleotides. Thus, for each gene (HA, NA, and NP) of the H1N1 and H7N9 subtypes, a pair of primers was selected that is highly conserved and specific to its subtype. As [the] additional criteria determining the suitability of the primers, the absence of duplexes and their annealing as well as the absence of erroneous annealing sites on the matrix of different strains were taken into account.

EXPRESSION OF HUMAN LACTOFERRIN IN TRANSGENIC TOMATO LINES ENHANCES THEIR RESISTANCE TO BACTERIAL AND FUNGAL PHYTOPATHOGENS

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Aim. Generation of transgenic tomato (*Lycopersicon esculentum Mill.*) lines of cvs. Money Maker and Lahidny expressing the *hLf* gene to enhance their resistance to phytopathogens. **Methods.** The agrobacterium-mediated transformation of tomato plants was carried out with the use of supervirulent strain EHA 105 carrying the pBin35LF plasmid with the *hLf* gene under control of CaMV 35S promoter and with the *nptII* gene conferring the resistance to kanamycin. The selection of transgenic tomato lines was carried out in the presence of 100 mg/l of kanamycin. Integration of [the] *hLf* gene into the genomes of the selected lines was confirmed with the use of PCR with the primers, specific to *hLf*. The expression of HLF was detected with the use of Western blotting with [the] monoclonal antibodies to lactoferrin. The resistance of transgenic tomato lines to *Ralstonia solanacearum* and *Clavibacter michiganensis subsp. michiganensis* was investigated using the disk diffusion assay, and to *Phytophthora infestans* – using the detached leaf assay and in vitro infection of transgenic plants. **Results.** In this study, the transgenic tomato lines with confirmed integration of *hLf* in their genomes were obtained. The expression of lactoferrin protein in transgenic lines was estimated at approximately 0.02-0.04% of total soluble protein. The growth inhibitory activity of the samples from transgenic plants expressing HLF on the bacterial pathogens (*C. michiganensis* and *R. solanacearum*) was shown using the disk diffusion assay. An increase in the resistance of the transgenic plants to *P. infestans* from 1 to 7 points of the 9-point scale was demonstrated using the in vitro infection of transgenic plants and the detached leaf assay. **Conclusions.** The obtained results show that the expression of human lactoferrin in transgenic tomato plants could enhance their resistance to bacterial and fungal phytopathogens.

DETECTION OF ZEARALENONE IN CEREALS USING SMARTPHONE-BASED MOLECULARLY IMPRINTED POLYMER MEMBRANE SENSOR SYSTEM

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Aim. The purpose of this study was to develop [an] advanced molecularly imprinted polymer (MIP) membrane-based sensor system for detection of a low-molecular-weight mycoestrogen zearalenone (ZON) in cereals using smartphone as a detector and quantifier, optimization of the composition of the MIP membranes, which were utilized in the proposed sensor as low-cost, highly sensitive elements for ZON recognition. **Methods.** The proposed sensor system is based on recognition properties of the ZON-specific MIP membranes, which were obtained using dummy template-based approach and synthesized with cyclododecyl-2,4dihydroxybenzoate as a template molecule and ethyleneglycolmethacrylate-phosphate as a functional monomer. The choice of functional monomer was based on the result of computational modeling. Triethyleneglycoldimethacrylate was used as a crosslinker. Smartphone (Meizu 16) equipped with [a] built-in high quality camera (20 MP, F/1,8) was used as a detector and quantifier of the response of fluorescent sensor associated with ZON bound to the MIP membrane surface. The sensor responses were registered by the smartphone camera 1 min after excitation by UV-irradiation, □=320 nm. The captured images were analyzed using smartphone application Spotxel® Reader (Sicasys Software GmbH, Germany). The ZON fluorescence intensity expressed in arbitrary units related to zearalenone concentration in the sample was received using smartphone application within real time. **Results.** We reported the advanced MIP membrane-based sensor system, which was used for detection of mycoestrogen zearalenone in cereals using smartphone. The MIP membrane-based smart sensor system showed low limits of ZON detection (1 μg/mL), whereas the linear detection range comprised 1-10 μg/mL. Selectivity of the developed MIP membranes towards ZON was investigated by using close structural analogues of zearalenone (zearalenol, 17-β-estradiol, bisphenol A, resorcinol). Furthermore, we successfully detected ZON in wheat, rye, and maize flour samples of different manufacturers, which suggest[s] that the proposed sensing method can potentially be applied to the on-site monitoring of mycotoxins in food. **Conclusions.** The low-cost and fast miniaturized instrument for the on-site monitoring and prevention of ZON contamination in food products and feeding stuffs was developed using MIP membrane as a sensing element and smartphone as a detector. Efficiency of the proposed MIP membrane-based sensor for ZON measurement in both model and real samples as compared to traditional analytical methods has been demonstrated. **Acknowledgement.** Financial support from Ministry of Education and Sciences of Ukraine and National Academy of Sciences of Ukraine.

IN VITRO SCREENING OF WHEAT (TRITICUM AESTIVUM L.) FOR TOLERANCE TO WATER DEFICIT

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Introduction. Bread wheat (*Triticum aestivum L.*) is an important crop grown worldwide on about 200 million ha in a range of environments, with an annual production of more than 600 million metric tons. Stress factors especially drought negatively affect the plant growth and development and cause a sharp decrease in the plant productivity. Using biotechnology is a promising direction in selection of drought-tolerant wheat genotypes. One of the screening techniques based on physiological traits is the use of mannitol to induce osmotic stress in plant tissues. **Aim.** Using low molecular mannitol, to conduct in vitro screening of winter wheat lines promising for the tolerance to water deficit. **Methods.** Winter bread wheat genotypes Liutestsens 37611, Liutestsens 60027, Liutestsens 37391, Erytrospermum 60025, Liutestsens 60049, Erytrospermum 60068, Liutestsens 60106, Erytrospermum 37326, Liutestsens 60100 and Erytrospermum 37003 were studied. Callus tissue was obtained from shoot apex explants of 3-day-old sterile seedlings. For callus induction the MS medium supplemented with 2,4-D in concentration of 2.0 mg/l was used. Mannitol in concentrations of 0.4, 0.6 and 0.8 M was used as a selective agent. Calli were planted in petri dishes (40 in each) in 4 replicates. In 4 weeks the calli were tested for [a percentage of live cells. To induce morphogenesis the calli were transferred to MS medium supplemented with 1 mg/l BAP and 0.5 mg/l IAA. In 3 weeks they were transferred to a modified medium for rooting. The rooted plants were transferred to *in vivo* conditions to produce seed generation. The obtained experimental data were processed by the methods of statistical analysis. **Results.** Genotypic responses to osmotic stress in the *in vitro* culture of wheat are manifested by different survival rate and different regenerative ability under the action of a stress factor. It was established that 0.6 M mannitol differentiates wheat genotypes for water deficit. It was found that the line Erytrospermum 60068 was the most resistant to osmotic stress because its calli under selective conditions had the highest survival rate and the highest regeneration potential. The regenerated plants were obtained only from the explants of this genotype after cultivation on the medium containing 0.8 M mannitol. The line Liutestsens 60100 was the most susceptible to water deficit because the mass necrosis and lack of regenerative ability were observed in its calli under selective conditions. From the induced calli the plant regenerants were obtained and their rearing, rooting and transfer to [the] *in vivo* conditions were optimized. **Conclusions.** The line Erytrospermum 60068 can be used as a valuable material for further breeding of wheat. The obtained results definitely contribute to the study of both theoretical and practical aspects of the wheat drought tolerance and can be used in further research as the elements of biotechnological and breeding programs.

PHYTOSTIMULATORY ACTIVITY OF PAENIBACILLUS POLYMYXA KB STRAIN

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Aim. One of the main means of increasing the productivity of crops is the use of physiologically active substances of microbial origin - phytohormones, vitamins, organic acids, exopolysaccharides, enzymes, antibiotic and antifungal compounds, etc. - in the technology of crop cultivation. A prerequisite for establishing the phytostimulatory activity of the *P. polymyxa* KB strain is the study of the spectrum of physiologically active substances – the products of *P. polymyxa* KB metabolism. **Methods.** The qualitative and quantitative determination of phytohormonal compounds was performed by high performance liquid chromatography. The antifungal activity of bacteria was determined by the method of counter cultures. **Results.** According to the results of the study of the auxin activity of *P. polymyxa* KB, it was found that a culture fluid (KF) at a dilution of 1:10 inhibits the rooting process in bean cuttings, which can be caused by high content of the physiologically active substances having an inhibitory effect on rhizogenesis. The greatest stimulating effect was at a dilution of 1: 100 - the total number of roots and their mass increased twice compared to the control. With an increase in the dilution rate of *P. polymyxa* KB, the stimulation of rhizogenesis decreased up to a dilution of 1: 1000. According to the results of quantitative analysis of the content of phytohormonal compounds in the supernatant of KF bacteria, it was found that the auxin content was 81.15 μg/g of dry biomass. Given that the use of IAA in high concentration always inhibits the growth of roots, and in low, on the contrary - contributes to their prolongation, based on the data obtained, we can assume that *P. polymyxa* KB synthesizes the optimal amount of extracellular auxins for plants, which is confirmed by the results of the biotesting of culture fluid on cuttings of beans, where this strain contributed to the process of rhizogenesis. It was first discovered that *P. polymyxa* KB produces abscisic acid - 9.36 μg/g of dry biomass, which can be important in the interaction of bacteria with plants, namely, to increase their resistance to abiotic stress and to induce plant resistance to phytopathogenic micromats. It has been established the ability of *P. polymyxa* KB to synthesize exopolysaccharides (amylopectin and levane), which testifies to the possibility of phosphate mineralization with their participation, as well as to potential protective properties. **Conclusion.** The physiologically active substances, synthesized by *P. polymyxa* KB can provide the stimulation of growth processes, resistance to abiotic stress and play an important role in protecting plants against pathogens.

FORMATION AND FUNCTIONING OF ENDOPHYTIC ASSOCIATION OF SOIL SAPROTROPHIC FUNGI CHAETOMIUM COCHLIODES WITH COMMON BUCKWHEAT

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Aim. Many representatives of soil saprotroph fungi can penetrate into the plant tissues forming endophytic associations with a positive effect on both micro- and macroorganisms. Thus the aim of our study was to establish the features of interaction between *Chaetomium cochliodes* 3250 and buckwheat plants. **Methods.** Conventional methods - biochemical (content of plant growth regulators, activity of succinate dehydrogenase, acid and alkaline phosphatase, exoglucanase, endoglucanase, β-glucosidase, polygalacturonase) and physicochemical (content of photosynthetic pigments, he total and active working root surfaces) - were used in the work. **Results.** We established the capability of *C. cochliodes* 3250 for the synthesis of growth-regulating substances. It was shown that *C. cochliodes* 3250 could produce 2.4-epibrassinolide, crucial for the plants' resistance to pathogens, and ergosterol. The synthesized indolyl-l,3-acetic acid and ergosterol can serve as the main mediator molecules in the process of formation of the *C. cochliodes* 3250 symbiotic systems with plants. We found the enzymatic activity of cellulase complex in the culture liquid of *C. cochliodes* 3250. Exogluconase activity in the fungus culture liquid was 0.67 units/ml of cultivation, meaning that *C. cochliodes* 3250 can degrade the cellulose crystalline state. Endogluconases ensure hydrolysis of amorphous cellulose to cellobiose (exogluconase activity of *C. cochliodes* 3250 was 0.52 units/ml). β-glucosidase completes the breakdown of cellulose and provides hydrolysis of celibiosis to glucose (β-glucosidase activity of *C. cochliodes* 3250 was 1.02 unit/ml). Polygalactorous activity of the culture liquid of *C. cochliodes* 3250 predetermines the process of depolymerization of the adherent layer of pectin between the adjacent walls of plant cells. The highest polygalacturonase activity recorded on the ninth day of cultivation of micromycete, was 2.95 units/ml. The pre-sowing treatment of buckwheat seeds by fungus caused the main physiological responses of plants typical for the formation of symbiosis: growth of the total (by 145 %) and active working (by 7 %) root surfaces, increased length of plants (by 27 %), increased area of leaves (by 11 %) and increased content of chlorophyll a and b (by 17 %). Moreover an increased activity of succinate dehydrogenase was observed in the roots of treated plants (by 1.5 times). We found that fungi increase the activity of acid (326 %) and alkaline (391 %) phosphatase in the roots of buckwheat during the entire vegetation period meaning that the formed symbiosis is effective. **Conclusions.** We established the ability of *C. cochliodes* 3250 to influence ongrowth-regulating substances, cellulose complex enzymes and synthesis of polygalacturonase. The pre-sowing treatment of buckwheat seeds by fungi was shown to activate the basic physiological responses of plants such as general adsorption and active working root surface, length and mass of plants, surface area of leaves, chlorophyll a and b content. All responces were higher.

IDENTIFICATION OF NOVEL DRUG TARGET FOR THE TREATMENT OF K. QUASIPNEUMONIAE ATCC 700603

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Motivation. Klebsiella species represent the group of widespread Gram-negative bacteria. Klebsiella strains may cause opportunistic infections including pneumonia, urinary tract infections, liver sepsis and bloodstream infections. For instance, *Klebsiella quasipneumoniae* gains attention due to its responsibility for infections with great morbidity and mortality. According to the research at Saitama Medical University Hospital and Saitama Medical University International Medical Center in 2014-2017 the mortality rate of *K. quasipneumoniae*-associated infections reached 11%. Recent studies demonstrated multidrug-resistance of the strain *K. quasipneumoniae* ATCC 700603 to antibiotics with oxymino-β-lactame group (ampicillin, cefpodoxime etc.). Taking into account these facts, the development of anti-*K. quasipneumoniae* drugs with novel mechanism of action is essential for decreasing the rate of mortality of *K. quasipneumoniae*-associated infections and preventing resistance of bacteria. **Aim.** The aim of the study was to find novel protein targets of *K. quasipneumoniae* ATCC 700603, predict their three-dimensional structure and characterize the binding sites of chosen molecular targets for further creation of bioactive compounds. **Methods.** Multistep bioinformatic proteome analysis was conducted to find the molecular targets, which were non-homologs to human proteome, essential for bacteria metabolic pathways and presented the target novelty. The sequence analysis was based on BLAST algorithms. Homology modelling was performed by web-server SWISS-MODEL. Identification and characterization of druggable pockets of three-dimensional structures of potential targets were based on PrankWeb and Discovery Studio tools. **Results.** *K. quasipneumoniae* proteome analysis resulted in the determination of two novel potential molecular targets of bacteria – type-1 fimbrial protein subunit A and phospholipid-binding lipoprotein MlaA. Druggable pockets of the receptor models were investigated and described for further search of antibiotics. **Conclusion**. This study reports the identification of two novel molecular drug targets using the proteome analysis of pathogenic strain *K. quasipneumoniae* ATCC 700603. These targets are non-homologs to human proteome, essential for bacteria survival and have subcellular or secondary membrane localization. The revealed druggable pockets for small organic compounds represent the chosen proteins as perspective molecular drug targets for the rational design of a compound with antimicrobial activity.

SPECTRAL STUDIES OF THE INTERACTION BETWEEN OLIGORIBONUCLEOTIDES AND ALCOHOL SUGARS

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Aim. Our previous studies have shown the possibility of oligoribonucleotides (ORNs) to form complexes with alcohol sugar D-mannitol (D-M). ORNs consist of ribonucleotide monophosphates. Therefore, we aimed to investigate [the] ORNs components potential to form [the] complexes with D-M and similar alcohol sugars. **Methods**. We studied the absorption spectra of pure aqueous solutions of four nucleosides (A, U, G, and C), their monophosphates and inosine, and polynucleosides sequences with the addition of four alcohol sugars (D-M, Sorbitol, Lactitol, Maltitol) separately. Also, we studied [the] fluorescence spectra of all these samples at room temperature. **Results**. The most significant changes in adenine derivatives with the addition of D-M were observed in 3D-fluorescence spectra. The addition of other sugar alcohols demonstrated similar spectral changes, but less significant than D-M. The weakest changes were associated with uridine derivatives. Inosine did not show any changes in spectra after the addition of all sugars. Comparing the fluorescence spectra of non-diluted and aqueous solutions with different concentrations of these samples and ORNs, we observed the formation of excimer-like complexes. Since nucleosides and alcohol sugars have disparate degrees of interaction, the specificity of the chemical structure influences their ability to form complexes. **Conclusions.** The complexation of ORNs with alcohol sugars also correlates at the level of their constituents. The two isomers: mannitol and sorbitol have different degrees of interaction. The level of interactions of nucleosides and their phosphates with alcohol sugars in descending order is A>G>C>U. Inosine did not show any changes after the addition of sugars.

DESIGN OF 5-HETERYLAMINO-1H-INDAZOLE DERIVATIVES AS NOVEL HUMAN PROTEIN KINASE CK2 INHIBITORS

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Motivation. To date, 66 drugs targeting protein kinases have been clinically approved. Protein kinases are the second most important group of drug targets, after G-protein-coupled receptors and major drug targets for oncology. Protein kinase CK2 (casein kinase 2) has attracted attention as a potential target for the treatment of cancer. Dysfunction of CK2 is associated with more than 20 types of cancer. There are no approved drugs yet that target this kinase. Therefore, the development of new compounds active toward protein kinase CK2 is in great demand. **Aim.** The aim of the study was to find novel effective inhibitors of human protein kinase CK2 among 5-heterylamino-1H-indazole derivatives. **Methods.** A combination of computer aided drug discovery and biochemical testing methods were used to achieve the main aim of this study. Molecular docking was conducted to predict binding mode and affinity of studied compounds to ATP-binding site of human protein kinase CK2. The CK2 direct kinase assay was utilized to test bioactivity of compounds using peptide substrate RRRDDDSDDD as a substrate. **Results.** Novel inhibitors of CK2 among 5-heterylamino-1H-indazole derivatives were found. The most active compound had IC_{50} value of 2 nM. The analysis of physicochemical properties showed a poor lipophilicity of the studied compounds. A series of new indazole derivatives was designed using SAR study of the compounds to improve their physicochemical properties; they were evaluated as CK2 inhibitors with molecular docking. **Conclusion**. Overall, our study suggests that 5-heterylamino-1H-indazoles are effective protein kinase CK2 inhibitors and have a great potential for further optimization and developing more potent inhibitors of CK2.

IN PLANTA TRANSFORMATION OF COMMON WHEAT (TRITICUM AESTIVUM L.) VARIETIES BY TPS1 AND TPS2 GENES

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Aim. Recently, the method of plant genetic transformation in planta became more popular comparing to Agrobacterium-mediated *in vitro* transformation and biolistic transformation because it is simple, cheap and does not require sterile conditions and using *in vitro* tissue culture. Trehalose is a non-reduced disaccharide that plays important role as stress protectant in many organisms including yeasts (*Saccharomyces cerevisiae*). The aim of the work was the construction of vectors with yeast genes of trehalose biosynthesis TPS1 and TPS2, Agrobacterium-mediated in planta transformation of wheat, and the study of selected transgenic plants on their tolerance to such osmotic stress, as drought. **Methods.** Wheat plants were transformed by *Agrobacterium tumefaciens* (strain GV3101) carrying vector constructions pBract214-TPS1 and pBract214-TPS2 with yeast (*Saccharomyces cerevisiae*) genes TPS1 and TPS2 of trehalose biosynthesis under constitutive maize ubiquitin promoter (PUbi) and selectable gene of hygromycin phosphotransferase (hpt). These constructions were created by Gateway-cloning technique. Several common Ukrainian wheat varieties were used for genetic transformation: Vykhovanka, Zhuravka Odeśka, Kesariya Polis′ka, Zymoyarka and Shchedrist′. In planta transformation was conducted using pre-neutered wheat spikes by standard method. Obtained seeds of wheat (transgenic generation T1) were germinated, and total DNA from the seedlings was extracted for PCR analysis in order to confirm the transformation events. **Results.** Integration and the presence of yeast genes in wheat genomic DNA isolated from transgenic plants were confirmed by PCR analysis using the primers specific to TPS1 and TPS2 genes. The estimated transformation efficiency for both constructions was around 3–5 %. **Conclusions**. According to the results of the work, yeast (*Saccharomyces cerevisiae*) trehalose biosynthesis genes (TPS1 and TPS2) were for the first time successfully transferred into common Ukrainian wheat varieties using Agrobacterium-mediated in planta transformation in order to enhance their drought tolerance.

ZNF217 PROTEIN EXPRESSION IN BCR-ABL-POSITIVE CHRONIC MYELOID LEUKEMIA CELLS

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Chronic myeloid leukemia (CML) is a myeloproliferative disease, the cytogenetic marker of which is the Philadelphia chromosome resulting from a reciprocal translocation between 9 and 22 chromosomes. The outcome of the formation of the Philadelphia chromosome is the expression of the Bcr-Abl oncoprotein. According to the results of mass spectrometry analysis, 23 proteins were identified as potential candidates for interaction with the PH domain of the Bcr-Abl oncoprotein. One of them is the zinc finger protein 217 (ZNF217), which acts as a transcription factor, promotes cell proliferation and counteracts apoptosis. Normal cells have a low expression level of ZNF217, whereas various oncopathology is characterized by its overexpression. **Aim.** The aim of the work was the establishment of colocalization of [the] Bcr-Abl/ZNF217 proteins and study of the relationship between [the] tyrosine kinase activity of Bcr-Abl oncoprotein and [the] ZNF217 protein expression in human chronic myeloid leukemia cells. **Methods.** K562 cells were cultured in RPMI 1640 medium with 10% FBS at +37°C and 5% CO_2. Immunofluorescence analysis was performed using anti-ZNF217 and anti-Bcr-Abl antibodies. The samples were visualized by confocal microscopy. Quantitative analysis of the results was performed using the Fiji software JACoP plugin. **Results.** Nuclear localization of ZNF217 in K562 cells was confirmed by immunofluorescence analysis. Colocalization of Bcr-Abl and ZNF217 in CML cells was established for the first time. Quantitative analysis of the results showed a positive correlation between Bcr-Abl/ZNF217 proteins, the Pearson correlation coefficient was r=0.524. According to Mander's overlap coefficients, which were 0.721 for M1 and 0.226 for M2, it may be concluded that there is a high overlap of localization signals of the target proteins. Here, it was shown that inhibition of the kinase activity of the Bcr-Abl oncoprotein led to low/or completely inhibited expression of ZNF217 in CML cells. **Conclusions.** It was demonstrated for the first time the colocalization of Bcr-Abl/ZNF217 proteins and interdependence between Bcr-Abl tyrosine kinase and ZNF217 protein expression in K562 cells. Thus, deregulation of the ZNF217 functions by Bcr-Abl could be one of the reasons for the uncontrolled cell proliferation and differentiation, genetic instability, and antiapoptotic response in the development and progression of CML.

SCREENING FOR SUBCLINICAL INFECTION OF THE MAMMARY GLAND OF COWS BY FLOW CYTOMETRY

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Aim. Subclinical mastitis in cows is a costly disease, [the] detection of which is difficult without special tests for diagnosis. The number of somatic cells (SC) in milk is used as an important indicator of udder health since SC are a part of the innate immune system. The aim of our study was to determine whether it is possible to identify subclinical mastitis in cattle at an early stage by a simple and fast flow cytometric method, then to presume the main cell populations in flow cytometric dot plots and, with these, to elaborate a method of mastitis prognostics. **Methods.** Samples of milk were collected from 20 cows. Centrifugation was performed to isolate cells from milk, and fat was removed using filter paper. Microscopy of cow milk cells was performed to exclude debris in the samples. After preparation, the milk cells were incubated with propidium iodide (PI), which differs between viable and non-viable cells. This procedure made it possible to localize cell types in a flow cytometry dot plot and to differentiate between viable and non-viable somatic cells. To show the percentage of cell apoptosis in milk samples, we used Annexin V-fluorescein isothiocyanate. **Results**. By means of the microscopic method, three samples above 150 000 (2/20) were identified. Our data of flow cytometry graphs (dot plot) showed that milk from cows with mastitis contained populations of cells accompanying inflammation (supposed macrophages, PMN), which could be detected by (FSS) and right angle signals (SS). During our study, we carried out the analysis with Annexin samples to distinguish pathological from physiological processes. It was shown that the number of apoptotic cells for pathological process in the mammary gland (1/20) was 42%, necrosis 53%, viable – 4%, milk samples (18) from healthy cows contained apoptosis cells 70%, necrosis – 11%, viable – 2%. Compared with sample SC-549 480 per 1 ml the percentage of apoptotic cells was 72%, necrotic – 8%, and viable – 10%. **Conclusions**. Somatic cell apoptosis allows increasingly assessing the state of immune homeostasis, which should be considered in immunology for the diagnosis of diseases. The determination of the number of events of the phagocyte secretion of the cows' mammary gland allows a more objective assessment of the state of local immunity of the udder [under] both normal and pathological conditions. The cell populations, present in milk, change during the development of inflammation and mastitis. A heterogeneous population of cells may also be present in cows with a subclinical course of mastitis. Flow cytometry screening of milk cells can serve as a diagnostic and prognostic criterion in monthly studies of all cows in the dairy herd to diagnose inflammation, and predict the development of subclinical mastitis. Proper monitoring of SC reduces the percentage of clinical outbreaks of mastitis and helps to avoid culling out milk and cows.

PARAMETERS OF LOCOMOTOR ACTIVITY, LIFE EXPECTANCY AND PRODUCTS OF LIPID PEROXIDATION IN SOD-MUTANTS OF DROSOPHILA MELANOGASTER UNDER THE ACTION OF SPERMIDINE.

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Aim. The goal of the study was to investigate the effect of spermidine on locomotor activity, life expectancy parameters and lipid peroxidation rates in *D. melanogaster* Sod-mutants. **Methods.** Control was descended from ebony / Oregon R crossing. Sod1 and e genes are localized in the third chromosome in Drosophila. In order to avoid the effects of e gene products on Sod1 gene expression we used heterozygotes of the test and control lines. The experiments were performed only on males. The effects of drug introduced into the medium at certain concentrations were analyzed by larval feeding. To determine the index of locomotor activity we used a climbing test with 13-15-day-old male of the control and experimental lines; the total mileage, washing time, movement time and rest time were determined by the «Open field» method. The content of the major lipid peroxidation (LP) products - diene conjugates (DC) and TBA-positive products, was determined by the Stalna method (Stalna et al., 1977). Total protein was determined by the Lowry method (Lowry et al., 1951). We performed statistical processing of the data in Microsoft Excel using the data analysis package, namely a two-sample t-test with different variances. [The] Survival curves were constructed using «GraphPad Prism 6». **Results.** Based on the determination of the locomotor activity index by the climbing test it was found a decrease of motor activity in Sod1-mutants to the level of control line over the drug's action. The index of locomotor activity in the Sod1-mutants without the influence of the drug was 0.326 whereas with the drug -]0.133; this decrease is significant (**). With the action of spermidine the total mileage decreased in both control and experimental individuals, with experimental flies having a statistically significant difference of this indicator compared to flies grown on standard medium; both control and experimental individuals significantly reduced movement time, more time insects were at rest and washing. We plotted the survival curves and analyzed the S50 under the action of drug. There was a significant ($p < 0.0001$) decrease in S50 for the action of spermidine in the experimental line by 40% and in the control by 28%. Intensification of lipid peroxidation as a result of oxidative stress is characteristic for Sod1-mutants that have impaired superoxide dismutase (an antioxidant balance enzyme). The main indicators of LP are DCs and TBAs, so we have found their level. Spermidine intensifies the processes of LP: in the control line [it] increases the level of DC (in the Sod-mutants remains unchanged), and in the experimental line – [of] TBA-products. **Conclusions.** The influence of spermidine is revealed as a decrease in the parameters of locomotor activity to the level of control line in Sod-mutants of *D. melanogaster*. The intensification of the processes of LP and reduction of the parameters of the average life expectancy were established for both lines of flies.

INTRACELLULAR LOCALIZATION OF BCR, ACTIN AND CLATHRIN DETERMINED BY FLUORESCENT MICROSCOPY

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Nowadays, chronic myelogenous leukemia (CML) is quite common type of cancer. A key pathogenetic factor in the development of this disease is the BCR/ABL protein expressed by the bcr/abl hybrid gene. Today, CML is treated with protein kinase inhibitors. However, this method leads to drug resistance and necessity to develop new generation of treatments. For this purpose, it is important to study the effect of BCR/ABL on intracellular signalling pathways and interactions. Previous studies have shown the BCR/ABL interaction with the cytoskeleton by ABL part, as well as the potential role of the PH domain of the BCR in actin reorganization. The cortactin is involved in the reorganization of actin during clathrin-mediated endocytosis, so there is also the possibility of BCR being involved in this process. The **aim** of this study was to determine a role of BCR part of the fusion protein in the reorganization of actin cytoskeleton by conducting fluorescence microscopy of K562 and HEK293T cell lines. **Methods.** Transfection of mammalian K562 and HEK293T cell lines was conducted with polyethyleneimine. Purified plasmids of pECFP-BCR, pm Cherry-clathrin for HEK293T cells and EGFP-clathrin for K562 cells were used for transfection. Transfected live HEK293T cells were stained with SiR-actin and transferred to a reusable aluminum microscope chamber. For transfected K562 cells, fixation and staining with mouse primary anti-BCR antibodies, ATTO647N-phalloidin, anti-mouse Alexa-555-conjugated secondary antibodies, and DAPI were performed. Next, fluorescence microscopy of live and fixed cells was performed using a Carl Zeiss LSM 510 Meta microscope. A 100x zoom lens with a 1.4 aperture was used. Image processing was done in Fiji. **Results.** Fluorescent microscopy images of live HEK293T cells and fixed K562 cells were obtained. Colocalization of BCR, actin, and clathrin protein was detected during microscopy of K562 fixed cells. Also, during microscopy of live HEK293T cells, actin branching and colocalization between actin, BCR and clathrin in the cytoplasm were observed. **Conclusions.** The results confirm that the BCR/ABL protein can affect the actin reorganization not only through the protein kinase function of the ABL part but also via the BCR part. Fluorescent microscopy of live HEK293Tcells transfected with empty backbone pECFP, pmCherry-clathrin, and stained with SiR-actin was performed as negative control, which confirms that the ECFP protein itself does not colocalize with actin and clathrin.

THE STUDY OF TRITICUM SPELTA L. IN VITRO REGENERATION

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Aim. To develop an effective methodology for in vitro culture regeneration of amphidiploid spelt wheat (Triticum spelta L.). **Methods.** For our experiments we used winter variety "Europe" of amphidiploid wheat (2n=42) spelt. It has been shown, that spelt regeneration occurred at callus-mediated stage. Mature embryos were used. Mature seeds were sterilized with 96% ethanol for 5 minutes, then 20 minutes with 5% NaClO, followed by three washings in sterile distilled water. For callus induction we used Murashige and Skoog medium (MS), supplemented with 2,4-D - 2 mg/l, AgNO3 - 10 mg/l and ceftriaxone - 250 mg/l. 21-day-old calli were obtained from mature embryos and transferred to regeneration media. We used MS medium supplemented with AgNO3- 10 mg/l, naphthaleneacetic acid (NAA) - 0.5 mg/l and 6-benzylaminopurine (6-BAP) - 2 mg/l. For regenerant plants we used medium containing half of salts and vitamins of MS medium with 2% sucrose and supplemented with AgNO3 - 10 mg/l and indole-3-butyric acid (IBA) - 0.7 mg/l. After the root system was formed, the regenerant plants were transplanted into the soil at greenhouse. We used PCR analysis for evaluation of genetic homogeneity and somaclonal variability of the R1 generation plants samples. We extracted genomic DNA from the leaves using CTAB method. PCR was conducted using primers to ISSR markers. The PCR fragments were separated in 1% agarose gel. The electrophoretic spectrum of water-soluble proteins was also analyzed. For this purpose, proteins were isolated from the crushed leaves using 1X phosphate buffer solution, with pH 7.4: 0.137 M NaCl, 0.0027 M KCl, 0.01 M Na_2HPO_4, 0.0018 M KH_2PO_4. The extracted proteins were separated in 30% acrylamide gel, according to the method proposed by Laemmli U. K. **Results.** The frequency of callusogenesis was 96.72%. The frequency of plants regeneration on MS medium supplemented with AgNO3 - 10 mg/l, NAA - 0.5 mg/l and 6-BAP - 2 mg/l was 29.32%. Effectiveness of rhizogenesis was 26.92%. The frequency of spike formation in rooted plant-regenerants was 28.57%. Regenerated plant comparison according to ISSR markers (Inter simple sequence repeats) and water-soluble proteins showed lack of somaclonal variability. **Conclusion.** In our study we showed ability of Triticum spelta L. to regenerate into in vitro culture. Spelt shows high callusogenesis frequency and normal regeneration frequency. Spelt regenerant plants did not show any somaclonal variability.

CYANINE DYES AS FLUORESCENCE PROBES FOR MICROSCOPY AND IN VITRO DETECTION OF PROTEINS AND NUCLEIC ACIDS

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Aim. In recent years, the fluorescent detection and visualization of the biomolecules, cell structures, and organelles are becoming increasingly popular methods of biomedical research. Thus, the aim of the research is to study cyanine dyes with different lengths of polymethine chain and substituents as fluorescent probes for proteins and nucleic acid *in vitro* detection and visualization in microscopy. **Methods**. Spectral-luminescent properties were characterized for unbound dyes and in the presence of biomolecules by fluorescent spectroscopy and UV-VIS absorption measurements. The visualization of stain cells was performed using fluorescence and confocal microscopy. **Results**. We have explored the series of monomethine cyanine dyes with benzothiazole and benzoxazole heterocycles and series of benzothiazole based pentamethine cyanine dyes for their fluorescence sensitivity to various biomolecules such as nucleic acids and some globular proteins. It was shown that the monomethine cyanine dyes are sensitive to nucleic acids. Fluorescence emission maxima for the dyes in the complex with nucleic acids lie between 470 and 529 nm. The highest emission increase by 480 times was observed for benzoxazole quinoline dye Sl-2598 in the presence of RNA. Obtained quantum yield value for this dye in the presence of DNA and RNA is equal to 40% and 54% respectively. Thus, we suggested these dyes to be sensitive to NA containing organelles, particularly to nuclei and nucleoli. Staining of live and fixed cells of the human breast cancer MCF-7 and human fibroblast cell line has shown [that] these dyes stain RNA-containing organelles – nucleoli and cytoplasm in live and fixed cells and are effective in quite low concentrations (1–5μM). The pentamethine cyanines are far red emitted dyes that show fluorescent sensitivity to serum albumins. We have observed that the addition of albumins leads to a significant increase in dyes fluorescence intensity. However, the fluorescent response of dyes in the presence of other globular proteins was noticeably lower. The value of fluorescence quantum yield for dye 1756Sl bearing sulfonate group complexed with HSA amounted to 42% compared with 0.2 % for the free dye. The detection limit of HSA by this dye was > 0.003 mg/ml that indicates the high sensitivity of dye to low HSA concentrations. According to molecular docking data, dyes could bind to up to five sites on the HSA molecule; the most preferable are the hemin-binding site in subdomain IB and the dye-binding site in the pocket between subdomains IA, IIA, and IIIA. **Conclusions**. Due to its high sensitivity, the monomethine cyanine dye Sl-2598 is suggested for further research as an efficient stain for nucleoli visualization in live and fixed cells in fluorescence microscopy. The pentamethine cyanine dyes 1756Sl could be proposed as powerful far-red fluorescent probe applicable for highly sensitive detection of albumins.

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INTERACTION OF INTERFERON A-2B WITH DIFFERENT FORMS OF OLIGONUCLEOTIDES

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Aim. Interactions of protein nucleic acids play a decisive role in many biological processes. RNA-based drugs that can bind and affect the work of epigenetic regulators and transcriptional proteins through interaction with regulatory domains, can be used as safe analogs. **Methods.** We studied the ability of yeast oligoribonucleotides (RNA), yeast RNANa salt (RNANa), and yeast RNA:D-mannitol complex (RNA:D-M) to affect the fluorescence quenching and conformational changes of Interferon α2b – a vital protein of the antiviral cell defence mechanism. To determine the energy parameters of protein ligand interactions, we used isothermal titration nanocalorimetry Nano ITC. **Results.** It is shown that the quenching curves of interferon fluorescence when interacting with salt forms of RNANa and RNANa:D-M coincide in the titration zone from 2 to 7 mkmol and have the same character in the range of 0.97 to 0.87 nInt. At the same time, the fluorescence quenching curve of this protein in the RNA shows significantly lower values from 0.93 nInt to 0.85 nInt in the titration range from 2 to 7 mkmol. These values are particularly strong in the study of the RNA:D-M, especially in the range from 1 to 4 mkmol, respectively, from 0.90 to 0.83 nInt. Thus, RNA, and especially RNA:D-M leads to a change in the conformational mobility of interferon α-2b by increasing the content of disordered regions. At the same time, salt analogues increase the number of structured secondary elements, such as α-helices, β turns and β antiparallel sheets and probably increase the conformational stiffness of interferon α-2b. The results of the study of enthalpy changes in the titration of interferon α-2b acid form of RNA and RNA:D-M was -63.28 kJ/mol and -96.61 kJ/mol, respectively, and for the RNANa and RNANa:D-M respectively 4.516 and 5.139 kJ/mol. The change in entropy when adding the RNA to interferon α-2b was -38.72 and in the case of the RNA:D-M -63.53 kJ/mol*K, . The change in entropy when adding the RNANa to interferon α-2b was 17.05 kJ/mol*K, and the RNANa:D-M, 17.58 kJ/mol*K. A similar pattern was demonstrated when studying the change in Gibbs energy during titration of interferon α-2b with RNA and RNA:D-M and it was -24.56 and -33.07 kJ/mol, respectively. And when titrated with RNANa and its RNANa:D-M, respectively -12.9 and -12.43 kJ/mol. **Conclusions.** These results of studying the effects of thermodynamics of different forms of RNA and their complexes with D-mannitol in the titration of interferon α-2b may indicate different sites of binding different forms of RNA to protein, as well as other modes of binding and various types of conformational changes in the protein.

BIOINFORMATIC IDENTIFICATION OF TYPE OF *THERMUS THERMOPHILUS TRANS*-EDITING FACTOR ALAX, ITS CLONING AND EXPRESSION

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Introduction. Accurate protein biosynthesis is an important aspect of cell life. It depends on different processes, e.g. the attachment of correct amino acids to cognate tRNAs by aminoacyl-tRNA synthetases (aaRSs) that together with additional *trans*-editing factors may prevent mistakes during translation. D-alanine is a structural component of bacterial cell walls, but is not included in proteins. Thus, the stereospecificity of aaRSs is essential for further proper translation. Previously, alanyl-tRNA synthetase was shown to misactivate $tRNA^{Ala}$ with D-alanine and be capable of editing D-Ala-$tRNA^{Ala}$ substrates. Possibility of *trans*-editing factor AlaX to correct such mistakes is unknown. Moreover, the stereospecificity of the AlaX family needs to be investigated. **Aim.** In this work we aimed to identify the type and conserved regions of the *T. thermophilus* AlaX, create the expression construct and test the expression level of target protein. **Methods.** NCBI database and BLAST were used to obtain *T. thermophilus* AlaX sequence and to search similar proteins. Sequences were aligned by the G-INS-i algorithm in the TREND tool. The type of AlaX was identified by analysis of conserved regions in resulted alignment and comparing the domain organization of the different types. Sequences of identified type from various organisms were aligned and analysed by the TREND tool. Target gene (1179 bp) was cloned into pET28b vector (NcoI/HindIII), transformed by electroporation into *E. coli* BL21(DE3) pLysS cells. The level of expression was analyzed in the following range of conditions: 1) induction by different IPTG concentrations (0.1, 0.5 and 1 mM); 2) various media (LB, TB, 2xTY, P); 3) various induction time (5 h and overnight, ON) and 4) temperature (+37ºC and +25ºC). Analytical purification on Ni-NTA resin was performed. **Results.** *T. thermophilus* AlaX was identified as AlaX-L type that consists of the editing and oligomerization domains. The Glycine-rich (N-domain) and Zinc-binding (C-domain) motifs and GG-motif (the oligomerization domain) are the conserved regions. The gene of *T. thermophilus* HB27 AlaX was cloned into vector pET28b and expression conditions of the corresponding protein in *E.coli* cells were studied. Suggested optimal conditions for AlaX expression were the following: TB media, +25ºC, ON and 0.5/1 mM IPTG. It was proved by analytical purification. **Conclusions.** AlaX type of *T. thermophilus* was identified as AlaX-L by analyzing conserved regions obtained by alignment and protein feature analysis. Selected optimal conditions of expression allow getting soluble protein and further optimization of purification procedure and functional studies of target enzyme.

THE STUDY OF NOVEL FUNCTIONALIZED MEROCYANINE DYES FOR ALBUMINS DETECTION

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Introduction. In the last decade, the development of far-red fluorescent probes for protein detection in solution and monitoring protein structure modification, particularly serum albumins remains relevant. Serum albumins are major transport proteins in blood plasma for many compounds like hormones, fatty acids, etc. A change in the level of serum albumin is one of the indicators of early pathological conditions, because it is highly linked to the general nutritional and inflammatory status. Methods of fluorescence measurements have attracted great attention due to their high analytical sensitivity, easy and fast implementation and relative simplicity. The development of fluorescent dyes that absorb and emit in the far-red region (650-980 nm) is highly applicable in many biological studies. These probes have certain advantages such as minimum photo-damaging of biological samples, high tissue penetration and allow imaging with minimal to no auto fluorescence. **Aim.** The main goals of our work are the synthesis and study of the spectral-luminescent properties of novel far-red fluorescent merocyanine dyes in the aqueous solution and in the presence of bovine serum albumin (BSA), human serum albumin (HSA), horse serum albumin (ESA), ovalbumin (OVA). **Results**. Novel series of merocyanine dyes containing modified polymethine bridges were synthesized and the spectral-luminescent properties of merocyanine dyes in aqueous solution and in the presence of BSA, HSA, ESA and OVA were investigated. The studied free dyes possess low to moderate fluorescence intensity in the aqueous buffer. The maxima of excitation spectra are located at 624-714 nm with the fluorescence emission maxima in the far-red area of the spectrum between 644-730 nm. The addition of serum albumins leads to the shift of excitation and emission maxima of the dyes to the long-wavelength spectral region for 8-12 nm that points to the binding of the dyes to proteins. The fluorescence intensity of all dyes increases in the presence of BSA, HSA and ESA. At the same time, this dye demonstrates noticeably lower fluorescence intensity in the presence of OVA, which is structurally different from serum albumins. **Conclusions.** Novel series of merocyanine dyes based on variations of heterocycles were shown to be fluorescent sensitive to serum albumins of similar structure (BSA, HSA and ESA) in contrast to OVA. The highest spectral response to albumins was shown, its fluorescence intensity increases by 48 times with BSA, by 64 times with HSA, and by 37 times with ESA. The novel series of merocyanine dyes are suggested as promising far-red probes for serum albumins detection. These dyes could be potentially applicable in fluorescent spectroscopy for protein detection and visualization with minimum to no auto fluorescence.

PURIFICATION AND CHARACTERIZATION OF HIGHLY SPECIFIC ANTI-COA ANTIBODIES

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Introduction. Coenzyme A (CoA) and its derivatives are involved in different metabolic pathways, and perform range of functions, including gene expression regulation and ATP biosynthesis. CoA molecule covalently binds to and modifies cysteine thiols of cellular proteins in response to metabolic or oxidative stress in a process called CoAlation. The main role of this novel post-translational modification is to protect proteins from over-oxidation that could lead to loss of function and protein degradation. Such redox-regulated modification was found to be widespread across prokaryotes and eukaryotes. The isoforms of ribosomal S6 kinase (S6K) were among the CoAlated proteins found. **Aim**. The goal of this work was to purify and test highly specific anti-CoA antibodies and to investigate the CoAlation modification of the set of human epithelial cell lines under metabolic and oxidative stress. **Methods**. In order to detect protein CoAlation, the unique in-house developed specific anti-CoA antibodies were purified and tested. Different human epithelial cell lines, and human embryonic kidney 293 (HEK293) cells stably overexpressing pantothenate kinase 1β (Pank1β) were used in the research. Metabolic stress of cells was induced via glucose and pyruvate starvation. To induce oxidative stress, cells were treated with 0.5 mM hydrogen peroxide and diamide (0.5 mM) for 30 min. Protein CoAlation was visualized using western blot analysis. **Results**. The unique monoclonal antibodies against CoA were purified and characterized. In western blot analysis of total cell lysates, the HEK293/Pank1β model cell line showed a slight level of protein modification under metabolic stress whereas a significantly higher level was shown for protein CoAlation during treatment with oxidising agents. Most of CoAlated forms occurred to be proteins with molecular weight of approximately 110 kDa, 85 kDa, 50 kDa. Using western blot analysis it was also demonstrated that the endogenous level of CoAlation of examined cell lines (HEK293, MCF7, Hep2G, HT29) under metabolic and oxidative stress was slightly observable, and not significant compared to the HEK293/Pank1β cells. **Conclusions**. Using the highly specific anti-CoA antibodies we, once again, confirmed that CoAlation modification appears as a protection from over-oxidation in HEK293/Pank1β model cell, and investigated the protein CoAlation level of various human cell lines under metabolic and oxidative stress.

INVESTIGATION OF CYTOTOXICITY AND EFFECTIVENESS OF POTENTIAL INHIBITORS OF MGMT IN HUMAN CELL CULTURES

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Aim. One of the reasons of low chemotherapy effectiveness is the increased activity of the reparative enzyme O^6-methylguanin-DNA methyltransfarase (MGMT) in tumor cells. That is why the inhibitors of MGMT are used in alkylating chemotherapy. O^6-benzylguanine is the most useful in practical medicine. Unfortunately, it and its known analogues revealed toxic activity at the third stage of clinical trials. So, this makes the search for alternative low-toxic and effective MGMT inhibitors actual and relevant task. **Methods.** 98 new potential inhibitors had been selected by the semi-flexible docking method. All of them were tested by biochemical method in uncellular conditions and a few of them were investigated by MTT test for toxicity with using cancer cell line Hep-2. The best nontoxic compounds were selected for the next step of the research with the clonogenic method, which is more sensitive, because it shows not only the cellular survival, but also the ability of individual cells to create clones, i.e. the presence of normal metabolism. The next step of the research was the study of the effectiveness of MGMT potential inhibitors in Hep-2 cells. In these experiments we also used MTT test and clonogenic method with an additional treatment of the cells with a model alkylating compound - nitrozoguanidine. Such a treatment creates the conditions similar to alkylating chemotherapy. **Results.** We selected 2 compounds (№41 and №59), which were not toxic at the dose of 10 mkg/ml and similar to O^6-benzylguanine in effectiveness if comparing their ability to increase the cytotoxic effect of nitrozoguanidine at low doses. Moreover, compound №41 revealed more expressed inhibitor activity than O^6-benzylguanine. **Conclusions.** In terms of toxicity and efficacy, 2 potential inhibitors have been selected from 98 tested compounds. Now we are going to test the inhibitors on animal models for their toxicity. It is also necessary to investigate their therapeutic effect in combination with alkylating drugs for chemotherapy on animal models.

EFFECT OF COMPLEXES OF OLIGORIBONUCLEOTIDES WITH D-MANNITOL ON THE VIABILITY OF DIFFERENT TUMOR CELL LINES

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Aim. Complexes of oligoribonucleotides with D-mannitol (ORNs-D-m), which are obtained by formation of complexes between the D-mannitol and total yeast RNA, have a wide range of biological activity. Previous studies on the murine B16 melanoma model have shown that solid tumor formation was not observed with the simultaneous administration of melanoma cells and ORNs-D-m. The aim of this work was to study the effect of ORNs-D-m on the viability of different tumor cell lines *in vitro*. **Materials and methods.** Two tumor cell lines: murine B16 melanoma and human U251 glioblastoma were used for the study and were cultivated under standard conditions. Oligoribonucleotides (ORNs), complexes of oligoribonucleotides with mannitol (ORNs-D-m), and their sodium salts were applied for the treatment of cells. The growth inhibition of tumor cells induced by these compounds was assessed with methyl thiazolyl tetrazolium (MTT). A comparison of the effect of compounds on cell viability was carried out by the concentration of half-maximal inhibition of IC50, which were determined based on dose-dependent curves received by using the GraphPad Prism 8.0.1 software. **Results.** The duration and dose of ORNs-D-m treatment affects on B16 viability. Thus, the IC50 was significantly reduced after 48-hours treatment compared to 12-hours treatment and was 2.7 ± 0.2 mg/ml. In contrast, the IC50 with ORNs treatment was in the range of 6.9 ± 0.4 mg/ml, which indicates their less pronounced effect compared to ORNs-D-m. For the U251 cell line, a similar trend in viability after treatment was found, with IC50 for ORNs-D-m – 1.2 ± 0.04 mg/ml, for ORNs - 10.1 ± 0.2 mg/ml. D-mannitol had no effect on cell viability. For comparison, salt forms showed either no inhibitory effect for B16 or effects at significantly higher concentrations of drugs compared to acidic forms for U251. **Conclusions.** ORNs-D-m inhibits the viability of both cell lines: murine B16 melanoma and human U251 glioblastoma. ORNs have a less inhibitory effect on viability. The acid form of ORNs-D-m has a more obvious effect on cell viability compared to the salt form.

TABLE OF CONTENTS

Alphabetical index