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Reviews

THE FORMATION OF BIOFILMS BY PSEUDOMONAS AERUGINOSA STRAINS, AND METHODS OF ITS CONTROL (REVIEW) 9-13
Sarkis-Ivanova V.V.

The majority of natural bacterial populations exist in the form of biofilms, highly-structured multicellular communities incorporated in extracellular polymer matrix of own production. For a considerable part of clinically significant species this form of existence provides optimal conditions for reaching of pathogenic and colonizational potential, and also encourages saving of metabolically inactive part of population which is characterized by a low level of sensitivity to antibiotics impact. The latter plays a significant role in formation of chronic persistent infections resistant to antibiotics treatment. Members of microbial group are united basing on the principle which excludes antagonism, determines their nutritional, energetic and another connections between them and environment. Such a connection of microorganisms community behavior received a special definition of "quorum sensing". Quorum sensing is an ability of some bacteria (probably, also another microorganisms) to communicate and coordinate their behavior through secretion of substances which are the signals for coordination of certain behavior or interaction between bacteria of the same type or subtype depending on their growth tightness. When the concentration of excreted signaling agents reaches a borderline value the group of bacteria starts working as a single organism. Furthermore, signaling agents for Gram-negative and Gram-positive microorganisms differ. For instance, for Pseudomonas aeruginosa microorganism a signaling molecule of quorum sensing is acyl homoserine lactone, and biofilm created by this pathogen produces pyocyanin which functions as an electronic transporter in current generation. A large-scale investigation of P.aeruginosa biofilm morphology, mechanisms of its production and degradation, peculiarities of formation under different conditions and impact of various factors are at the infancy stage. According to undivided opinion of scientists who investigate this problem the main source of nosocomial diseases and persistence factor of their causative agents in hospital ecosystems from air and water to inner surfaces of catheters and body systems are represented by biofilms. Bacteria of P. aeruginosa type are human opportunistic pathogens which being a part of biofilm may cause different nosological forms of pyoinflammatory diseases with severe course and high fatality in immunocompromised patients. These microorganisms are able to contaminate external and internal surfaces of catheters, probes, respiratory tubes, lenses, and form biofilm on them. The given information determines reasonability of searching the methods of control of biofilm production in P. aeruginosa cultures which can be used to increase the effectiveness of antibiotic treatment in blue pus infection and/or decontamination of medical equipment, and another objects of hospital environment. One of the methods to control biofilms produced by P. aeruginosa on abiotic surfaces from different materials is impact of antiseptics and decontaminants. Modern decontaminants are represented by a wide spectrum of chemical compounds of different classes which have a common ability to destroy microorganisms even in low concentrations. At present the impact of bacteria on separate elements of biofilm formation is considered to be one of the most perspective targets for the action of new antimicrobial medicines. Numerous investigations of recent 40 years demonstrated that the process of biofilm formation is complex and multistage. Currently the principal directions of development of new antimicrobial preparations are the following: development of antiadhesive coatings and preparations able to interrupt the function of eternal structures of cells in charge of adhesion (fimbriae, adhesins); development of preparations that block synthesis or destabilize matrix of biofilm; development of preparations that cause cell adhesion deficiency in microcolony, and block cell fission; development of preparations that block intercellular signaling (quorum sensing), and cause disconnection of cells from biofilm and their transition into plankton existence or resist pathogenicity factor expression. Nowadays the influence of certain physical factors on biological properties of biofilms is studied as well. At the present time the new technology that is photodynamic therapy is in intensive progress. Low intensity electromagnetic radiation has widespread application practically in all areas of medicine. In fact, under the influence of light emitting diode radiation metabolic and functional properties of biological system variety can be significantly changed. According to a number of investigators a direct method of light emitting diode radiation effect provides direct influence on cellular structure elements, moreover it has been proved that membrane structures of cell are the most sensitive to optical radiation effect. It was found that under influence of light emitting diode radiation a breakdown of daily biofilms with inhibition of plankton cells production capacity was observed. All the above allows to assess current problem of medical science and practice that is process of biofilm formation in microorganisms particularly in Pseudomonas aeruginosa strains. Analysis of literature sources shows practicability of research line of specific subject that is proved by crisis of antibiotic therapy observed for now and characterized not only by multitude of resistant microorganisms but also by absence of preparations and infection pathology therapeutic regimen that have assuring effectiveness and therefore search for alternative control methods is critical and challenging.
Keywords: Pseudomonas aeruginosa, formation of biofilms, antibiotics, physical factors, photodynamic therapy

PROSPECTS FOR USE OF CONDENSED GASES AND SUPERCRITICAL FLUIDS IN PHYTOCHEMICAL PRODUCTION 14-21
Demyanenko D. V., Breusova S. V., Karpenko L. A.

In the given review article analysis of the literature and patent sources concerning main methods for intensification of extraction processes of medicinal vegetative raw materials – use of condensed gases and supercritical fluids (SCF) on more acceptable extractants has been carried out for last 20 years. Urgency of the specified technologies consists in need for replacement of traditional extraction methods on power- and time-saving ones, and also in use of nontoxic, fire-proof and low-boiling solvents because the most of routine organic solvents (ethanol, methanol, acetone, chloroform, ethylacetate, etc.) are toxic and/or flammable or expansive and rather hard to evaporate out from extracts obtained. The abovementioned trends are the most universal for intensification of extraction processes and sometimes purification of final or intermediate products acceptable for commercial scale of manufacture. The main advantages and disadvantages of the given methods are compared for different plant species and groups of biologically active substances (BAS). It has been shown that in most cases supercritical CO_2 (SC-CO_2) are inferior in its dissolving ability to number of condensed gases and, besides, such technology is much more expensive. The range of BAS taken with SC-CO_2 is limited to mainly lipophilic compounds because of zero electrical dipole moment of SC-CO_2 and its low polarity. As extractants alternative to SC-CO_2 with higher dissolving

ability SC - ethane, nitrogen monoxide, freons - R134a, R23, R32, R408 and number of others can be used. Also to enlarge range of extractable BAS it is possible to add different cosolvents, mainly ethanol or methanol in quantity up to 20%. At the same time in phytochemical production prospective alternatives to liquid or supercritical CO_2 are certain condensed gases with wider range of physico-chemical properties: fluorinated derivatives of hydrocarbons (freons), liquid ammonia, dimethyl ether (DME), sulfur hexafluoride (insulating gas) or their mixtures, etc. Their major characteristics include lower vapor pressure if compared with liquid CO_2, antimicrobic activity allowing to solve one the main problem in phytochemical production – microbial contamination of extracts (and other herbal drug preparations), possibility to extract not only lipophilic, but also more polar substances depending on choice of solvents or their mixes and their higher extraction rate.It has been found that some kinds of freons (for example, R22) due to their higher polarity are able to take wider spectrum of BAS than liquid CO_2: essential and fat oils, fat-soluble vitamins, coumarins, carotenoids, phenolic alcohols, valrates, iridoids, some alkaloids and flavonoids. Besides, certain freons (for example, C318) have very high selectivity allowing to extract essential oils without accompanying fats. Some condensed gases, such as liquid ammonia, dimethyl ether and difluoromethane (freon R32) can be used as well for obtaining of hydrophilic BAS (saponins, alkaloids, flavonoids). Thus such solvents should be polar enough or can be mixed with polar modifiers. Supercritical fluids and some subcritical condensed gases are suitable for fractionating of primary (crude) extracts because their selectivity considerably depends on temperature, pressure and composition (in case of mixtures with each other or with cosolvents). Also high selectivity of condensed gas and SCFs is shown in near-critical areas. Very important property of most of condensed gases and SCFs is their ability to considerably reduce microbial contamination of extracts in comparison with initial plant raw materials. **Conclusions.** Among existing methods for intensification of stage of plant extraction the most applicable for commercial scale is use of condensed gases and supercritical fluids as extractants. It is found that for today in the world the most widespread SCF is carbon dioxide. The main lacks of CO_2 as an extractant are high working pressure and narrow spectrum of extractable BAS which is limited only to lipophilic substances. This induces the search for alternative condensed gases and SCF among which fluorohydrocarbons (freons) are of the greatest interest. Also perspective methods of extraction intensification are applications of ultrasound, microwave field and some other methods of raw material processing, but meanwhile they have not got industrial value because of insufficient scientific base.
Keywords: condensed gases, supercritical fluids, plant raw materials, extraction, freons, carbon dioxide.

Experimental papers

STANDARD TREATMENT REGIMENS FOR MULTIDRUG-RESISTANT TUBERCULOSIS – ANALYSIS OF EFFECTIVENESS 22-27

Ovcharenko I. A.

Introduction. In recent years, the number of cases of multidrug-resistant tuberculosis (MDR-TB) has been increasing in Ukraine and all over the world. The leading factor in fight against this pathology is the effective treatment of such patients. To increase the effectiveness of treatment of patients with MRI in 2008 unified protocols for the provision of medical care to patients were introduced in Ukraine, which offered standard treatment regimens for such patients, taking into account the individual sensitivity of pathogen to antituberculous drugs. **Aim**. Analysis of effectiveness of various standard therapy regimens that were used in patients with newly diagnosed pulmonary MDR-TB. **Materials and methods**. 68 case histories of patients with newly diagnosed pulmonary MDR-TB who were treated in the Kharkov Regional TB Dispensary № 1 in 2009-2014 and received anti-tuberculosis therapy according to current clinical protocols for medical care to patients with chemoresistant TB. All patients were older than 18 years. Patients were divided into two groups. Group I included 49 people who received treatment according to the Order of Ukrainian Ministry of Health № 600 of October 22, 2008. "Standard of medical aid for patients with chemorreistant tuberculosis". Patients from this group received the following standard therapy regimen: 6EZAm(Km)QEt(Pt) / 12-18EZQEt(Pt); 6EZAm(Km)QPAS / 12-18EZQPAS. In addition, 19 patients (38.8%) in group I were treated with isoniazid (H). Thus, in group I there were 2 subgroups: subgroups Ia (with the use of H) - 19 patients and Ib (without using H) - 30 patients, and the effectiveness of treatment in them was analyzed. Another 19 people, group II, received therapy according to the Order of Ukrainian Ministry of Health № 1091 of December 21, 2012. "On approval and introduction of medical and technological documents for standardization of care in tuberculosis" according to the scheme 8ZKm(Am)LfxPt(Et)Cs(Tz,PAS) / 12ZLfxPt(Et)Cs(Tz,PAS). All patients underwent clinical tests (fever, cough, body weight deficit), x-ray (lung destruction, X-ray dynamics) and bacteriological (pathogen detection by smear and cultureon solid and/or liquid media) signs of pulmonary tuberculosis at 2 and 6 months from the beginning of chemotherapy. **Results and discussion**. After 6 months of treatment, in group II, fewer patients complained of cough (15.8% and 44.9% in groups II and I, respectively); presence of destruction was noted in 47.4% and 67.3% of patients in groups II and I, respectively, indicating healing of cavities in a larger number of patients from group II. Effectiveness of treatment of patients in both groups was almost the same (69.4% and 68.4% in group I and II, respectively), but in the group I there were by 5.9% more deaths, and treatment failure by 4.4% than in group II. To identify possible causes of such differences, we analyzed within group I of subgroups Ia and Ib. Healing of cavities was noted in larger number of patients in group Ib, because the destruction in 6 months of treatment was determined in 56.6% and 84.2% of patients from the subgroups Ib and Ia, respectively. Also, after 6 months of treatment there were more patients with bacterioexcretion detected by both smear microscopy (10.5% and 0%) and culture (21% and 10%) in subgroup Ia than in subgroup Ib, respectively. Effectiveness of treatment was almost the same (68.4% and 70%) in subgroups Ia and Ib, respectively. There were more deaths in subgroup Ib by 18.1% than in group Ia (5.2% and 23.3%, respectively). When comparing groups of patients treated without isoniazid (groups II and Ib), it was established that in group II, after six months of treatment there were by 27.5% patients with cough (15.8% and 43.3% respectively) less than in a smaller patient than in subgroup Ib; also, after 6 months of therapy in this group, there were 9.2% less patients with destruction of lung tissue (47.4% and 56.6% in groups II and Ib, respectively). In addition, at the 2nd month of treatment there was positive X-ray dynamics in more by 10.9% patients in group II than in group Ib (84.2% and 73.3%, respectively). Also, after 2 months of treatment, there were less patients with bacterial excision (detected both microscopically (26.3% and 30%, respectively), and culturally (42.1% and 60%, respectively)) in group II than in group Ib. Effectiveness of treatment of such patients was almost the same (70% and 68,4%, respectively, in groups II and Ib), but the number of deaths was higher in group Ib than in group II (23.3% and 10.5%, respectively). Also in group II there were no treatment failures. **Conclusions**. Analysis of treatment efficiency criteria for patients with MDR-TB in various standard schemes showed that in group II the schemes were most effective. It should be noted that expressed positive effect of therapy has been already was achieved at the 6th month of treatment, despite the fact that according to the protocol, duration of intensive phase was not less than 8 months. But the additional use of isoniazid had no positive effects, and in general, when comparing the effectiveness of treatment with regimens with its use, patients from this group showed the worst positive dynamics from therapy.
Keywords: Multidrug-resistant tuberculosis, treatment effectiveness, criteria of treatment effectiveness, standard treatment regimen

SURVIVAL OF MICROORGANISMS FROM MODERN PROBIOTICS IN MODEL CONDITIONS OF THE INTESTINE 28-33

Kabluchko T.V., Bomko T.V., Nosalskaya T.N., Martynov A.V., Osolodchenko T.P.

Introduction. The staye of intestinal microflora affects the work of the whole organism. When composition of normal ibtestine microflora changes, its restoration is required. In our days a wide variety of probiotic drugs are available on the market which can be used to solve this problem. Most bacteria having probiotic properties represent the families *Lactobacillus* and *Bifidobacterium,* which have poor resistance to acidic content of the stomach and toxic effects of bile salts. Various studies have clearly shown that in a person with normal acidic and bile secretion, the lactobacilli and bifidobacteria are not detected after the passage through the duodenum, i.e., they perish before reaching the small intestines. In this study we compared the survival of different microorganisms which are contained in 9 probiotic drugs in a model of gastric and intestinal environments. **Material and methods.** In the laboratory of SI: "Mechnikov Institute Microbiology and Immunology, National Ukrainian Academy Medical Sciences" the *in vitro* experiments have been evaluated to test the ability of different probiotic bacteria which were contained in 9 probiotic drugs to survive the impact of the model environment of the stomach and duodenum. *Bacillus coagulans* persistence was evaluated under impact of simulated environment of the stomach and duodenum, it also was assessed by the quantity of CFU by incubation on culture medium. The following were studied: *Lactobacillus acidophilus, Lactobacillus rhamnosus, Lactobacillus reuteri, Lactobacillus casei, Lactobacillus plantarum, Lactobacillus bulgaricus, Bifidobacterium bifidum, Bifidobacterium longum , Bifidobacterium breve, Bifidobacterium infantis, Bifidobacterium animalis subsp. Lactis BB-12, Saccharomyces boulardii, Bacillus coagulans, Bacillus clausii, Enterococcus faecium.* Microorganisms were incubated for 3 hours in a model environment of the stomach (pepsin 3 g / l, hydrochloric acid of 160 mmol / l, pH 2.3), later after centrifugation and washing, they were incubated for 3 hours in intestinal model environment (bile salts 3% pancreatin 0.1%, pH 7.0). Inoculation was performed before incubation, after incubation in the gastric medium and after incubation in intestinal medium. We used the medium corresponding to the studied genus of bacteria - MRS-environment for lactobacilli, bifidum for *Bifidobacterium,* sabouraud medium for the isolation of yeasts and fungi and endo agar for the isolation of *Enterobacteriaceae*. We assessed the quantity of CFU before and after impact. **Results and discussion.** After incubation in a simulated gastric environment, bacteria of the type *Lactobacillus* and *Bifidobacterium* did not survive and were not defined. Only *Bacillus coagulans* and *Saccharomyces boulardii* were resistant. These microorganisms grew after incubation in the same amount as before incubation - 10^{5-6} and 10^{7-8} CFU respectively. *Bacillus clausii* also survived in these conditions, but to a lesser extent: initially - 10^7 CFU, after incubation - 10^5 CFU. After staying in model environment of the duodenum *Bacillus coagulans* and *Saccharomyces boulardii* were still fully viable, and the number of germinating *Bacillus clausii* bacteria decreased by an order - up to 10^4 CFU. **Conclusion.** The probiotics containing *Bacillus coagulans* and *Saccharomyces boulardii* showed complete resistance to the impact of the model environment of the stomach and duodenum, *Bacillus clausii* was partially resistant. It leads to conclusion that probiotic drugs containing lactobacilli and bifidobacteria, cannot withstand the aggressive environmental influence of the stomach and duodenum and become inactivated under their influence. Probiotic drugs Enterol containing yeast *Saccharomyces boulardii*, and Laktovit Forte containing the spore-forming bacterium *Bacillus coagulans* are completely resistant to the action of the model environment of the stomach and duodenum.
Keywords: probiotics, intestine, stability to acids and bile.

NONTUBERCULOUS MYCOBACTERIOSES: EPIDEMIOLOGY, CLINIC AND POSSIBILITIES OF LABORATORY DIAGNOSTICS IN MODERN CONDITIONS **34-38**

Shevchenko O.S., Novohatska M.F., Sharapova O.V, Pogorelova O.O.

Introduction. Today there are more than 150 species of nontuberculous mycobacteria (NTMB) known, of which 99 were registered in Europe. Due to the similar clinical picture with tuberculosis, mycobacterioses are difficult to diagnose and often occur with TB as a mixed infection. Also, there are cases of NTMB detection in the sputum of persons previously suffered with tuberculosis, which can lead to a false diagnosis of recurrent disease. Currently, the role of atypical mycobacteria in human pathology is increasing. Atypical mycobacteria are characterized by a wide spectrum of sustainability and potential pathogenicity to humans and animals. It is generally accepted that the reservoir of infection are animals and the environment (water, soil). NTMB presence in water can lead to the erroneous diagnosis of mycobacteriosis due to laboratory contamination of samples with nontuberculous mycobacteria from the outside. Moreover recently it has been found the possibility of transferring NTMB from person to person on the background of existing lung disease. For many types of infections reservoir still has not been found. Aim of this work was to study identified in the Kharkiv region cases of non-tuberculous mycobacterioses, features of their clinical manifestations and laboratory diagnostic options. Materials and Methods. We examined 32 patients (25 men and 7 women), residents of Kharkiv and Kharkiv region, Ukraine, who were diagnosed with "non-tuberculous mycobacteriosis of lungs" during 2014-2016. Patients were examined with routine diagnostic algorithm for TB. Nontuberculous mycobacteriosis was diagnosed on the basis of NTMB growth in BACTEC system, after which the diagnosis was verified by the following criteria: 1. Smear: the absence of formation of Cord-factor (in the smear NTMB are located scattering); 2. Negative immunoassay (ID-test); 3. Negative GeneXpert MTB/RIF Then non-tuberculous mycobacteria were identificated by investigation on solid media. They were divided on groups of susceptible causative agents according to Runnyon classification and the modified classification, approved by the Order Ministry of Health of Ukraine №45 from 06.02.2002, which takes into account the NTMB growth rate, character and color of colonies. Results and discussion. In all 32 reported cases infiltrative changes were identified radiographically. In 13 patients (40.6%) cavities were found. In addition in smear of 10 patients (31.2%) was found scant bacterial excretion, 2 (6.2%) - moderate, and 1 (3.1%) - massive. According to the results of culture NTMB were identified. Chromogenic slow-growing non-tuberculous mycobecteria prevailed (65,4%). Another causative agents belonged to M. avium complex. The heaviest lesions were caused by chromogenic slow-growing non-tuberculous mycobecteria, as 41,1% of these patients had extensive lung damage, and 64,7%% - destruction of lung tissue. Radiographic changes correlated with the severity of clinical manifestations. Also in this group most often was found detectable by smear bacterial excretion, including moderate and massive. In our patients prevailed cavernous form which is is typical for smoking middle-aged men, often develops on the background of previous lung disease - cystic fibrosis (M. abscessus), COPD, bronchoectatic disease, tuberculosis in the past, aspergillosis (M. xenopi), as well as on the background of HIV infection and drug treatment that suppress the immune system (corticosteroids, TNF-alpha inhibitors). Clinical and radiographic this form is almost indistinguishable from pulmonary tuberculosis (infiltrates in upper lobes with formation of cavities), more severe, often with a syndrome of intoxication, cough, shortness of breath, sometimes hemoptysis. Without treatment, the disease progresses rapidly and leads to formation of large cavities in the lungs and respiratory failure. The most common pathogens that cause such clinical and radiological picture are M. kansasii, M. xenopi and Mycobacterium avium complex. Conclusions. In Kharkiv region there is an increase of non-tuberculous mycobacterioses, but their diagnostics is difficult due to the lack of diagnostic capabilities: it is impossible to perform CT examination in all patients, no possibility of reliable identification of the type of pathogen (no DNA strip technology, tablet technology, DNA sequencing, high-performance liquid chromatography). Among the identified cases of mycobacterioses prevailed chromogenic slow-growing NTMB. Clinical and radiographic pattern was characterized by severe thoracic (cough, shortness of breath) and intoxication (fever, weight loss, weakness) complaints and extensive lesions of pulmonary system. Also in this group was often found detectable by smear bacterial excretion, including massive one. It was noted that often mycobacterioses developed on the background of existing pulmonary disease. Failure to conduct complete laboratory diagnostics, identification of pathogen and determination of its individual sensitivity to drugs significantly complicates prescribing of adequate chemotherapy regimens. In this

regard, we consider a priority the development of standards for diagnosis and treatment of non-tuberculous mycobacterioses and further strengthening the capacity of laboratory diagnostics.
Keywords: non-tuberculous mycobacteria, epidemiology, diagnostics

MOLECULAR BIOLOGICAL RESEARCH AT FATAL CONSEQUENCES OF VIRAL MYOCARDITIS 39-43

Smelyanskaya M. V., Peremot S. D., Kashpur N. V., Volanskiy A. Y.

Introduction. Diagnosis of viral myocarditis, based on the evidence base, is still one of the key problems of the heart disease. The presence of morphological features of the inflammatory process makes it possible to confirm the diagnosis of myocarditis, but, at the same time, the absence of these features is not sufficient to remove this diagnosis. In routine postmortem study of deaths in multidisciplinary (non-infectious) hospital myocarditis is stated as a cause of death in 0.2-0.4% of all the autopsies. Mortality in myocarditis depends on the severity of the underlying disease, premorbid background, age and sex composition of the patients. According to different authors, it is very different and ranges from 0.03 to 26%. The aim of the work was to carry out histological and molecular biological studies postmortem material for confirming the etiologic role of herpesviruses with fatal consequences of infectious myocarditis during the observation period 2015-2016 years. Material & methods. The material of pathological heart, vascular endothelium, nerve ganglia, kidneys, liver and pancreas were investigated. Viral antigen detection was performed by fluorescent antibody technique with specific sera labeled with FITC (Dako Corporation, Carpinteria, CA) and detection of the viral genome by PCR (in SYNEVO Laboratory). Morphological studies have been conducted in the post-mortem offices of the Kharkov clinical hospitals. Detection of viral genome was performed by PCR using certified commercial kits for detection of nucleotide sequences of herpesviruses «HSV I, II-EPh», «VZV-FL», «EBV-EPh», «CMV-EPh», «HHV VI-Eph», («AmpliSens»). Diagnosis was made in «real time» using modern six-channel thermocycler «Rotor Gene 6000» (Qiagen, Germany). The first group consisted of 19 people who died from infectious myocarditis (group 1). The second group (group 2) consisted of 22 dead from complications of other cardiovascular disease. Pathoanatomical material of 11 people was used as a control group. Death in this group occurred as a result of traumatic injuries. The average age of those groups was 31 ± 3,8 years. All groups were matched by sex and age. Results & discussion. It has been found, that DNA HSV1,2 turned out in infectious myocarditis group and in the group with the same frequency cardiovascular disease. Whereas DNA HHV6 and CMV appeared in infectious myocarditis group 6-7 times more often. Noteworthy the DNA VZV finding of a significant percentage of myocardial tissue samples of the dead from group 1 in relation to other groups. According to our data, which coincides with the tendency of foreign research, the proportion of finding enteroviruses, compared with herpesviruses, is insignificant in all the groups. In all of the dead of the main group (infectious myocarditis) herpes viruses have been detected in several organs simultaneously. Also infarction hypertension virus is most often in the liver, pancreas and nerve ganglia. Thus, when HSV was expected in the nerve ganglia, kidney and vascular endothelium, the detection of a relatively high percentage of hypertension HHV6, CMV and, especially, VZV was quite unexpected in the pancreas and liver tissues. Detection of different herpes viruses in various organs confirms the pantropism of viruses of this family. And the persistence of the virus in one of the bodies makes it possible to its participation in the etiopathogenesis of infectious myocarditis. Conclusion. In deceased patients with viral myocarditis HHV6 and CMV, VZV and EBV are most often found in the myocardial tissue. In all of the dead of the main group (infectious myocarditis) herpes viruses have been detected in several organs simultaneously. Also infarction hypertension virus is most often in the liver, pancreas and nerve ganglia. In the group with infectious myocarditis 3-4 viruses and 5 or more viruses appeared significantly more often in comparison with the group with cardiovascular disease in two and 4,5 times, respectively. This may increase the virulence of the various representatives of herpesviruses from general cell receptors.
Keywords: herpesviruses, myocarditis, PCR

ORGANIZATIONAL AND LEGAL RESEARCH OF INDICATORS OF INCIDENCE AND PREVALENCE OF DIABETES MELLITUS IN COUNTRYSIDE AREAS 44-53

Zbrozhek S. I., Shapovalova V. A., Shapovalov V. V., Hmelevskiy N. A.

Introduction. For recent decades in Ukraine populations' health indicators became quite serious negative trends: increased mortality rates among people of working age, morbidity, reduced life expectancy and more. It should also be noted that one of the main indicators of the state, its civilization and competitiveness is the degree of orientation of the state to improve the health of citizens, legal and organizational improvement of the healthcare sector. The financial and economic crisis has significantly exacerbated the problems associated with the provision of medical care in Ukraine, ensuring its availability and good quality. The current healthcare system is fair and complaints from the public, and the medical staff, it is still not able to adequately meet the needs of the population in healthcare, to ensure the availability, quality and timeliness of health services, adequate prevention of morbidity, mortality and more. The need for healthcare reform emphasizes the failure to ensure the proper conditions existing in state funding and to implement effective schemes for full competition in the healthcare and pharmaceutical provision in countryside areas is therefore, in Ukraine implemented new organizational and legal approaches in reforming the health service. That is why in Ukraine implementing processes that are transparent, will reduce unnecessary costs for public administration and improve accessibility of the medications in countryside areas of different clinical and pharmacological, classification, nomenclature, legal and regulatory groups used in the pharmacotherapy of diabetes mellitus. **Materials and methods.** The organizational and legal, forensic and pharmaceutical researches were conducted in countryside areas at the regional level on example of the Kharkiv region on the basis of 62 public healthcare institutions, 16 public enterprises of healthcare and health companies of other ownership. Materials of the research were: the legal framework for the organization of pharmacy of the healthcare system in countryside areas; forensic and pharmaceutical practice concerning the complaints on countryside accessibility for their antidiabetic drugs; regional statistics of incidence and prevalence of diabetes mellitus. In conducting the research used the following methods: legal, documentary, bibliography, comparative, forensic and pharmaceutical, graphical analysis. **Results and discussion.** In the limited funding of the healthcare system and low pharmaceutical ensuring of patients with diabetes mellitus in countryside areas, the question of the optimal use of funds to avoid negative consequences, as evidenced presented in the article on an example of forensic and pharmaceutical practice. Among the possible reasons for such organizations to ensure pharmaceutical provision for privileged categories of citizens can point to the constant rise in prices for antidiabetic drugs and delay in timely registration of wholesale prices for these drugs. Also during the study was a comparative analysis of incidence and prevalence of diabetes at the regional level by the example of Kharkov region. The increase in the prevalence of diabetes is due to various factors, including stress, obesity, aging population, quality of food and life. The increase in the prevalence of the disease may indicate a lack of financing health systems, insufficient qualifications and training of medical personnel, the remoteness of rural health facilities and inadequate organization providing pharmaceutical rural antidiabetic drugs passivity farmers regarding preventive examinations, treatment their health, compliance with recommendations and a low level in the chain of relations "doctor - patient with diabetes mellitus - pharmacist." **Conclusions.** Diabetes mellitus treatment should be based on the principles of pharmaceutical law provided in Art. 4 of the Law of Ukraine "On the basis of legislation of Ukraine on healthcare" from the state budget. Also in reforming of the healthcare system must take into account the experience of the European Union, and the need to propose such changes to the legislation: - twice increased

funding by providing targeted subsidies from the state budget; - Statement of the Cabinet of Ministers of Ukraine of 02.07.2014 p. Number 240 "On reference of pricing for medicines and medical supplies procured for the state and local budgets"; - Annual review, update and adjust the list of antidiabetic drugs domestically who can buy HCF regional level in rural areas, fully or partially financed from state and local budgets approved by the Cabinet of Ministers of Ukraine dated 05.09.1996, number 1071 .
Keywords: diabetes mellitus, incidence, prevalence, circulation, antidiabetic medicines, pharmacy, forensic pharmacy, pharmaceutical law.

SYRUP AMKESOL REDUCES THE LEVEL OF PRIMARY AND SECONDARY PRODUCTS OF LIPID PEROXIDATION AND TOXIC METABOLITES OF NITRIC OXIDE IN BLOOD SERUM OF IMMATURE RATS WITH BRONCHIOLITIS **54-61**

Storozhenko K.V.

Aim of this study was to determine the pharmacological effect of syrup Amkesol evaluating the blood serum levels of primary and secondary products of lipid peroxidation, activity of AO enzymes and final rates of NO metabolites in immature rats of different ages with experimental model of bronchoalveolitis. Materials and Methods: The study was carried out on 90 WAG immature rats of ages 1, 2 and 3 menthes, that correlates by morpho-functional features to 4, 10 and 14 years of human age respectively, on a model of bronchoalveolitis. Experimental animals in each age series were randomly divided into 5 groups (n = 6): intact (healthy), 2 groups of control (untreated with bronchoalveolitis) 7 and 14 days, and two groups with bronchoalveolatis that received S-AKS daily during 7 and 14 days. The pathological process implemented by inhalation of irritant (Sephadex A-25 Pharmacia, Sweden (5 mg/kg)). In the blood serum samples activity of CAT, SOD, content of DC, TBARS, total NO, nitrates and nitrites were determined. Probability of the results was evaluated by using GraphPad Prism Software. The critical level of significance was taken equal to 0.05. Results. The use of S-AKS on the 7th day in the group of 1- and 3-month-old rats significantly exceeded activity of CAT compared to the control group of animals of corresponded age. The SOD activity in group of 2-month-old animals was restored to intact level, the activity of CAT exceeded the baseline level and amounted to 129% (P≤0,05). The concentration of DC in 1-month-old rats was lower by 45.3% in 2 monthly - by 12.5% than in the control group, in the group of 3 month-old animals - restored to normal (P≤0,05). The level of NO metabolites was significantly decreased compared to the corresponding control group in all age series (P≤0,05). After 14 days of treatment with S-AKS in all age group of animals observed restore the contents of primary and secondary products of lipid peroxidation compared to the intact level. Conclusion. Administration of S-AKS reduced the values of primary and secondary products of lipid peroxidation; restored the activity of antioxidant enzymes; reduced the level of NO and its stable metabolites to the physiological level in all studied age groups of animals with experimental bronchoalveolitis.
Keywords: children's dosage forms, syrup Amkesol, bronhoalveolitis, prooxidant-antioxidant system, nitric oxide metabolites.

PHARMACOLOGICAL RESEARCH OF THE DENTAL GEL WITH CARBON DIOXIDE HUMULUS LUPULUS EXTRACT **62-66**

Melnik A. L., Kazmirchuk V. V., Nosalska T. M., Bomko T. V., Dovga I. M., Povolokina I. V., Kuchma I.Y., Martynov A. V.

Introduction. In contemporary pharmaceutical practice among drugs used in dental gels are most effective, that are easily applied to the mucous and long held on the gums to form a protective film and prolonging therapeutic effect. Therefore it developed a new drug with a carbon dioxide Humulus Lupulus extract antimicrobial action in gel form for treatment of the oral cavity. The aim of our study was to investigate the acute and subacute toxicity the gel containing 0.5 % carbon dioxide Humulus lupulus extract. The toxicity of the gel studied at the intragastric route of administration, due to the scope of drug – dentistry. Materials and methods. The object of the research was gel containing 0.5% carbon dioxide Humulus lupulus extract. Studies of acute toxicity gel with carbon dioxide Humulus lupulus extract was performed on white inbred laboratory mice of both sexes, weighing 19–21 gram. Animals received gel single intragastric at maximum tolerated this route of administration dose – 2.0 g/kg [6]. The experiment used by 5 mice of both sexes. The criteria of judgment about the toxicity was the clinical picture of intoxication, animal survival, dynamics of body weight of mice (raw data, 3, 7, 14 days). Observation of animals were carried out within two weeks. Subacute toxicity studied on white inbred laboratory rats of both sexes, weighing 220 – 250 g. Animals were divided into 3 groups, each experimental group were 5 male and 5 female rats. Total experiment used 30 rats. Rats was injected gel with carbon dioxide Humulus lupulus extract once a day for 14 days at doses of 0.2 g/kg and 1.0 g/kg (1/10 and 1/2 of the maximum dose in acute experiment). Control animals were injected comparison drug Kamistad–H gel at a dose of 1.0 g/kg. Evaluation of the toxic effects of investigational gel and reference medicine on the body of the experimental animals were carried out on the following parameters: clinical observations, the survival of animals, food and water consumption, the dynamics of body weight, hematological and biochemical parameters of blood. Clinical observations of the animals was performed daily during the experiment, registering changes in their general condition, behavior, food and water consumption. Clinical and biochemical parameters studied using a reagent kit of "Felicity Diagnostics" (Ukraine) and conventional methods. Body weight of animals recorded in dynamics. The animals were weighed before the experiment, then 7 and 14 days. Results and discussion. Results of the study parameters acute toxicity gel with carbon dioxide Humulus lupulus extract showed that intragastric administration to mice at a dose of 2.0 g/kg caused no clinical symptoms of intoxication. Animals calmly reacted to the introduction of the sample gel, their general condition and behavior remained unchanged. Animal deaths and abnormalities in their general condition and behavior during the observation period were noted. Impact analysis of the sample of gel on the dynamics of body mass animal experiments showed that a single injection of a gel to mice at a dose of 2.0 g/kg did not significantly affect the dynamics of body weight. In experiments on rats were studied subacute toxicity gel with carbon dioxide Humulus lupulus extract. The results of 14 days intragastric introduction gel and reference drug studies at subacute toxicity showed that daily administration of no effect on the general condition, appearance (state of the skin and mucous membranes) and the behavior of rats. Consumption of food and water in animal experimental and control groups had no differences from each other. During the experiment animal deaths were not reported. Data characterizing the dynamics of increase in body weight of rats showed that the animals of experimental groups and the control group by the end of the experiment had equivalent, statistically significant weight gain compared with the output data: males – $p \leq 0.0001$ females – from $p \leq 0.0001$ to $p \leq 0.0004$. During the experiment, the animals of all experimental groups were observed physiological fluctuations that characterize the pattern of peripheral blood. Results of biochemical studies showed that overall the tested parameters: the concentration of total protein, albumin, thymol test, the activity of alanine and aspartate aminotransferase, the concentration of glucose and cholesterol in the blood serum of rats of all experimental groups answered normal values. There have been fluctuations in biochemical parameters in animals of all experimental groups compared with baseline data did not go beyond the physiological norm.
Conclusions. 1. Study of acute toxicity after a single intragastric administration to mice gel with 0.5 % carbon dioxide Humulus lupulus extract have shown that gel does not cause death of animals; no toxic effects on the general state, behavior, food consumption and water weight animals; does not affect the absolute and relative weight of internal organs; does not cause visible changes of internal organs. 2. According to the standard classification gel with 0.5 % carbon dioxide Humulus lupulus extract refers to practically non-toxic substances. 3. Subacute toxicity study results showed that the gel with 0.5 % carbon dioxide Humulus lupulus extract with 14 daily

intragastric administration to rats at doses of 0.5 g/kg and 1.0 g/kg, does not cause death of animals; has no toxic effects on the general state, behavior and dynamics of body weight of the experimental animals; no negative impact on the performance of blood of animals; does not change the biochemical parameters characterizing the functional state of the liver of animals. 4. Pharmacological studies of dental gel with 0.5 % carbon dioxide Humulus lupulus extract, such as the results of the study of acute and subacute toxicity found that designed gel to include low-toxic substances (IV class of toxicity), has good tolerability and safety of long-term use.
Keywords: gel with carbon dioxide Humulus lupulus extract, acute and subacute toxicity, laboratory animals, clinical observations, reference drug – Kamistad-Gel H

THE PHARMACOLOGICAL EFFICACY OF THE EXTEMPORANEOUS GEL DIAVENOL **67-70**

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Introduction. Diavenol - new extemporaneous gel for the treatment of venous ulcers. The gel contains a synergistic combination *C. majus* quinolizidine alkaloids and riboflavin. Activate tissue regeneration shown previously for riboflavin, and cheleretrin / sanguirythrine. The combination of the alkaloids and riboflavin pharmacologically fully justified. The research aim was to check the mechanism action of the Diavenol gel - through stimulation of the pluripotent cells growth and determination the effectiveness of the gel for wound healing at experimental model in rats. **Materials and methods.** In a study 128 rats were used, of which 38 - in the pharmacological experiment with a stencil wound healing, and 90 in the experiment to study the influence of gel on CD34 cells amount in the blood in rats with chemically induced immunodeficiency. High immunodeficiency caused a five-fold administration of cyclophosphamide, prednisone and rubomycin. CD34 expression level was determined using flow cytometry FACS Calibur. **Results and discussion.** Topical skin application the Diavenol gel samples in rats helped to accelerate the regeneration process is about 2 times, as evidenced by the excess of blood parameters in experimental group animals to the 30th day, more than 2 times, while the indicators in the control group animals came to normal values only on the 60th day. Increasing the number of pluripotent cells as well as acceleration of wound regeneration indicates preferential stimulation of stem cell growth in the periphery, rather than an enhancement of bone marrow function. The maximum effect of the Diavenol observed on the 60th day and the restoration of the pluripotent cells physiological level have been observed on the 10th day after the start of Diavenol application. Due to the fact, that the gel was applied to intact skin, we can say that Diavenol had resorptive properties. Treatment of wounds in rats Diavenol gel resulted in a more rapid removal of inflammation and significant acceleration of stencil wound healing. In terms of the granulation appearance and wounds epithelialization the Diavenol gel has some advantage in relation to the dexpanthenol ointment.
Keywords: diavenol, CD34 + cells, regeneration, wound healing, riboflavin, Chelidonium majus, chelidonine, chelerythrine, sangviritrin.

ANTIBACTERIAL AND ANTIOXIDANT PROPERTIES OF SOME SELECTED EGYPTIAN PLANTS **71-95**

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Introduction. Medicinal plants have been used as a source of therapies since ancient times in Egypt. The present study was designed to investigate the anti-bacterial and anti-oxidant activity of different extracts from 20 selected medicinal plants of Egypt. Materials and methods. The disk diffusion method followed by microbroth dilution were used to determine minimum inhibitory concentration of the plant extracts against 10 bacterial strains belonging to 5 species, Pseudomonas aeruginosa, Klebsiella pneumonia, Escherichia coli, Staphylococcus aureus, Streptococcus pyogenes. While qualitative phytochemical screening followed by DPPH (1,1-diphenyl-2-picrylhydrazyl) assay were used to assess the anti-oxidant of the extracts. Results. The results indicated that all studied crude extracts were able to inhibit the growth of at least three of the tested bacteria. Moreover, all studied plants have various bioactive phyto-chemicals and were observed to be high to moderate antioxidant agents. Conclusion. Finally, the target of this paper is to describe the most interesting plant extracts investigated here to be alternative medicines.
Keywords: Anti-bacterial, Anti-oxidant, Medicinal plants, Phyto-chemicals, DPPH.

ANTIMICROBIAL ACTIVITY OF THE SUBSTANCES RECEIVED FROM RAW MATERIALS OF *LAMIACEAE* AND *CUCURBITACEAE* FAMILY PLANTS **96-100**

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Introduction. The search of new sources of raw materials for medicine creation is one of the pharmacy priorities. In this, our attention was drawn by the plants which are widely cultivated in Ukraine and have a sufficient source of raw materials. It is summer squash *(Cucurbita pepo L.)* and sowing cucumber *(Cucumis sativus L.)* of *Cucurbitaceae* family and also *peppermint* (*Mentha piperita L.)* of two sorts "Chernolistaya" *and* "Zgadka" of *Lamiaceae* family. However the drugs with antimicrobial action derived from the leaves of these plants in the market of Ukraine are absent. Spirit, lipophilic and polysaccharidic fractions were received from the leaves of sowing cucumber, summer squash and two sorts of peppermint by us. Antimicrobial activity studying of the received substances derived from the studied raw materials for expansion of the medicine range is promising. The relevance of researches is that getting for the first time due to complex raw material processing the lipophilic, spirit and polysaccharidic fractions of leaves of a sowing cucumber, summer squash and two sorts of peppermint "Chernolistnaya" and "Zgadka" are systemically studied on existence of antimicrobic activity. **The purpose of our research** is the studying of antimicrobial activity of getting substances derived from the leaves of sowing cucumber, summer squash and two sorts of peppermint "Chernolistnaya" and "Zgadka". **Materials and methods.** Raw materials for substances are the leaves of a sowing cucumber prepared in June, 2015 in the Kharkiv region, the village Selection, in the conditions of industrial cultivation (the selection station of Institute of vegetable-growing and melon-growing of UAAN). The leaves of summer squash were being prepared also in June, 2015 in Valkovsky district of the Kharkiv region, the settlement Dobropolye. The leaves of peppermint of "Chernolistnaya" and "Zgadka" sort were prepared on the industrial squares of Dnipropetrovsk region, Sofiyevskiy district, the settlement Ordo-Vasilyevka. Lipophilic fractions were received in Sokslet's device. As an extractant, chloroform was used. From the received extraction the extractant was being deleted in vacuum. Lipophilic fraction represented viscous substances of black-green color with typical fragrant. For receiving spirit fractions derived from solvent cake after receiving the lipophilic fractions it was used a method of fractional maceration at the general ratio raw materials-extractant 1:15, as an extractant was 50% ethyl alcohol. Extraction was carried out three times. The integrated extracts were concentrated in vacuum. After a full removal of an extractant and drying, it had been received the hygroscopic powders of green-brown color with specific tastes and smells. From the solvent cake which was remained after receiving lipophilic and spirit fractions polysaccharidic fractions representing water-soluble polysaccharides were evolved. They were friable powders of light gray color. The lipophilic fraction of sowing cucumber leaves, lipophilic fraction of summer squash leaves, lipophilic fraction of peppermint leaves of "Chernolistnaya" sort, lipophilic fraction of peppermint leaves of "Zgadka" sort, spirit fraction of cucumber leaves, spirit fraction of summer squash leaves, spirit fraction of peppermint leaves of "Chernolistnaya" sort, spirit fraction of peppermint leaves of Zgadka sort, polysaccharidic fraction of cucumber leaves, polysaccharidic fraction of summer squash

leaves were being studied. Test of antimicrobial effect of substances was carried out by method of serial dilution concerning the following six reference test cultures: *Staphylococcus aureus* of ATCC 6538-P, *Candida albicans* of ATCC 885-653, *Escherichia coli* of ATCC 25922, *Bacillus subtilis* of ATCC 6833, *Bacillus cereus* of ATCC 10702, *Pseudomonas aeruginosa* of ATCC 9027, according to the State Pharmacopoeia of Ukraine of 1 edition, in the Department of microbiology and immunology of KMAPQE. **Results and discussion**. The obtained data demonstrate that the widest range of antimicrobial activity was shown by SSL, ChPL and SPZ in the ratio of *E. coli*, *C. albicans* and *P. aeruginosa*. Thus, SSL, ChPL, SC and SPZ had a bactericidal activity in dilution 1:10 and 1:100 concerning *P. aeruginosa* and *E. coli*. PZL - concerning *P. aeruginosa*. SSL, SC, SSS, ChPL, PZL, SPCh and SPZ have antimycotic activity concerning *C. albicans*. Concerning *E. coli* it was revealed that SSL, ChPL, SPZ had bactericidal activity, and concerning *P. aeruginosa* and *C. albicans* SC, ChPL, PZL, SPZ had it in dilution 1:10 and 1:100. Among the polysaccharidic fractions the antimicrobic activity was revealed only for PC in both dilution concerning *B.cereus*. As for *S. aureus* and *B. subtilis*, bacteria of these strains were resistant to all studied fractions. **Conclusions.** Antimicrobial activity of a number of substances of sowing cucumber leaves, summer squash and two sorts of peppermint "Chernolistnaya" and "Zgadka" is studied. Lipophilic fractions of raw materials (except CL) have an antimycotic activity concerning *C. albicans.* Among all the polysaccharidic fractions only PC have antimicrobial action concerning *B. cereus.* Strains of bacteria *S. aureus* and *B. subtilis* are resistant to both dilutions of all studied substances.
Key words: antimicrobial activity, leaves, cucumber, summer squash, peppermint, substance.

EFFICIENCY METHODS OF PHYSICAL REHABILITATION OF OSTEOCHONDROSIS OF THE LUMBAR SPINE **101-103**

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Introduction. Osteochondrosis – the most common disease of the spine, which affects more than 70% of the population. Important is the social significance of osteochondrosis, in the structure of diseases of the musculoskeletal system as a whole the average primary disability of adult consequence of osteochondrosis – 1,7 to 10 000. It is important finding the most effective methods of complex treatment, and most importantly rehabilitation and recovery of patients with osteochondrosis. **Material & methods.** We selected 16 patients with osteochondrosis of the lumbar spine, men aged 29-42 years with disease duration from 2 to 9 years. To determine the effectiveness of the methods of physical rehabilitation of patients with lumbar osteochondrosis outside the period of exacerbation assessed the dynamics of three indicators: vertebral syndrome, extravertebral syndrome, subjective assessment of the general condition of patients by psychological testing. After the examination, the patients divided into two groups of 8 people. Patients of the first group received physical therapy (ultrasound therapy and massage therapy). Pharmacotherapy is not done. Patients of the second group received medications according to the recommendations received after discharge from hospital. **Results & discussion.** After the rehabilitation treatment severity of vertebral syndrome significantly decreased to 1,1±0,23 points ($p < 0,05$) in first group and to до 1,0±0,33 points in second group ($p < 0,05$) with no significant difference between groups of patients. These figures suggest that all patients there was a reduction in pain to a weak pain, but some patients even before its termination. After rehabilitation terms of pain rate in the first group significantly decreased to 1,1±0,03 ($p <0,05$) points; in the second group, the figure dropped significantly to 1,2±0,08 ($p <0, 05$) points. In patients who did not receive pharmacotherapy, improvement in psychological testing "HAM" was slightly higher; reflecting the more positive assessment by patients of their condition by the end of the course of rehabilitation, reduce fatigue, increase stamina, optimism, activity and energy. In the group first patients, unlike the second groups reducing the intensity of pain occurred against the background of physiotherapy simultaneously with a reduction in anxiety. **Conclusions.** Thus, even an isolated application of physical rehabilitation of patients with spinal osteochondrosis is effective and does not always require pharmacological support.
Keywords: osteochondrosis rehabilitation, conventional methods, efficiency