

2018 № 2

Contents

Editorial Board

Contents

Review

IMMUNOPROPHYLAXIS OF PSEUDOMONOSIS: ACHIEVEMENTS AND PERSPECTIVES

C.
(P.)
1
2-4

5-15

Gorodnitskaya N.I., Gabysheva L.N., Derkach S.A., Martynov A.V.

The review article presents a retrospective analysis of attempts to develop effective vaccines for pseudomonas prophylaxis. *P. aeruginosa* vaccine has been sought for 40 years; however it is still not available. The increased understanding of *P. aeruginosa* pathogenesis and its virulence factors supported the recognition of potential immunogens and passive immunotherapy that could be used for the development of an effective vaccine. These immunogens are situated in structural components such as lipopolysaccharides, pili, flagella, outer membrane proteins or are part of secreted products such as proteases, exotoxins and mucoid exopolysaccharides. There have been significant advances in later years; nonetheless there is clear need for additional basic research to further increase the understanding of those elements of immune response to *P. aeruginosa*. Recently, the antigenic drift of the *P. aeruginosa* actual strains has changed significantly, and no one has studied such changes in Ukraine, despite the high mortality from pseudomonas in the intensive care unit. The development of a multi-strain vaccine based on the actual strains of *P. Aeruginosa*, in our opinion, can significantly reduce the formation of nosocomial circulation of multidrug-resistant strains, and contribute to reducing morbidity and mortality rates from nosocomial pseudomonosis.

Keywords: pseudomonosis, vaccines, development, protective properties

Experimental works

A STUDY OF ANTIMICROBIAL ACTIVITY OF FOAM-WASHING AGENT SPECIMENS AT ACIDIC PH VALUES

Strilets O. P., Petrovska L. S., Baranova I.I., Bespala Yu. O.

Introduction. It is well-known that any parapharmaceutical substance, in particular, foam-washing agents comprising water in combination with detergents, extracts, water-soluble vitamins, viscosity regulators, pH, etc., is the ideal environment for microbial growth. Therefore, it is indispensable to use preservatives to protect any foam-washing agent from possible contamination by microorganisms. The modern trend is to use multicomponent preserving agents. Their main advantages are: presence of a single antimicrobial and antifungal effect, expanded range of effects, decrease in the risk of resistance of microorganisms and decrease in the toxicity and concentration of the preserving mixture. In this regard, the shelf life of parapharmaceutical substances is not provided through the use of large quantities of preservatives, but thanks to their rational combination. **Materials and Methods.** For this study, we have made a number of samples of foam washing bases with a number of preservatives, which are often used in developing foam-washing agents with acidic pH value, namely: sample number 1 – foam washing base + sodium benzoate; sample number 2 – foam washing base + «Euxyl K300» (phenoxyethanol, methylparaben, butylparaben, ethylparaben, propylparaben, isobutylparaben); sample number 3 – foam washing base + «Germaben II» (polypropylene glycol, diazolum dinomovine, methylparaben, propylparaben); sample number 4 – foam washing base + «Nipaquard CMB» (benzyl alcohol, triethylene glycol, chloromethylisothiazoline, methylisothiazoline). The concentration of preservative in each sample was 0.1% (average concentration that is recommended for developing foam-washing agents). The antimicrobial activity of prototype gels was studied in vitro by diffusion in agar ("wells" method). The antimicrobial activity was measured immediately after sample preparation. All the studies were performed in aseptic conditions using a laminar box (biological safety cabinet AS2-4E1 "Esco" Indonesia). **Results.** According to the study, it was found that among the selected preservatives "Nipaquard CMB" was just the most active. When studying the antimicrobial activity of foam-washing agent samples with different concentrations of the preservative "Nipaquard CMB", it was found that namely the sample with the concentration of "Nipaquard CMB" of 0.1% showed satisfactory results due to its antimicrobial activity against all cultures such as bacteria and fungi. **Conclusions.** On the basis of microbiological studies it has been demonstrated that all the selected preservatives such as sodium benzoate, "EuxylK300", "Germaben II" and "Nipaquard CMB" at a concentration of 0.1% have a broad spectrum of antimicrobial action and antimicrobial activity against all test strains used. We just chose «Nipaquard CMB» as a preservative at a concentration of 0.1% according to the results of experimental research, because it had the best results and a very high antimicrobial activity both against the bacterial cultures - *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, *Pr. Vulgaris* and in relation to the effect on fungal cultures – *Candida albicans*, *Aspergillus brasiliensis*.

Keywords: biological researches, preservative, antimicrobial activity, foam- washing agent, pH value.

INFORMATION TECHNOLOGIES IN CLINICAL AND PHARMACEUTICAL MANAGEMENT OF NEWBORN PATIENTS WITH ROTAVIRUS INFECTION: RETROSPECTIVE ANALYSIS

Soloviov S O., Kovaliuk O.V., Leleka M.V., Ivanov M.O., Dzyublyk I.V.

Aim. A retrospective analysis of the effectiveness of pharmacotherapy of newborn patients with rotavirus infection using the developed information technology. **Materials and Methods.** We proposed to evaluate therapy outcome based on survival analysis approach. Since cohort of patients has two available states: alive or dead, we proposed to use two states: hospitalized and discharged from hospital. Therefore, therapy effectiveness was associated with durations of stay in hospital. For a certain cohort such effectiveness could be displayed as probability curve of stay in hospital, so a lower curve reflexes higher intervention effectiveness. This could be graphically described as transmission of patients in a cohort between two states: hospitalized and discharged patients. **Results.** This approach was based on the developed computer program "Clinical and Pharmaceutical Management of Viral Infections" and tested with the use of 85 medical records for newborn babies from 5 to 60 days of life born in the period from 2001 to 2002 and came from maternity hospitals to the 1st and 2nd outbreaks of newborns and the intensive care unit of the NHSL "OKHMATDIT" mainly in a difficult condition: with clinical manifestations of gastrointestinal disorders, hypoxic or hypoxic-hemorrhagic lesions of the central nervous system, hyperbaric bilirubinemia, respiratory

distress syndrome, etc. Analysis of medical records showed that all hospitalized patients were examined for RVI by the presence of rotavirus antigens in the clinical material (feces) by the indirect hemagglutination reaction method, the most accessible and widespread at that time in the laboratory. The principle of the method was that pretreated formalin or tannin erythrocytes (more often human or sheep), on the surface of which the specific antibodies are sorted, in the presence of a homologous antigen form aggregates, manifested by the phenomenon of agglutination. Among the patients studied proved positive 60 persons (70.6%), 32 of whom received basic pathogenetic therapy. The analysis showed that the onset of positive therapy outcomes for patients with RVI was longer, and therefore, it is more likely to remain in the hospital for the first 1 to 20 days of the disease. In a detailed analysis, both in all patients and only in patients with RVI, it was noted that the severity of the leading symptoms at the end of therapy was significantly reduced. However, the use of complex therapy with $\alpha 2b$ -interferon was characterized by faster reverse development of clinical manifestations of the disease than in patients who did not receive interferon. In comparison, it was also investigated the use of smectites - anti-diarrheal agents of natural or synthetic origin with sorption properties. The obtained results gave a confident ability to indicate their effectiveness, which was determined by a significant decrease in the probability curve of hospitalization days, both in the analysis of data of all patients and only in patients with RVI. **Conclusion.** The retrospective analysis using the developed IT showed that rectal application of recombinant $\alpha 2b$ -interferon and smectites in the complex therapy of RVI in newborns can increase the clinical efficacy of therapy, namely, positively affect clinical manifestations of the disease through more rapid elimination of a number of symptoms. The results of the study prove that ICT based on pharmacoeconomic modeling can become an effective tool for clinical and pharmaceutical management of patients in a hospital, and is a reliable source for assessing the recovery rate, which is necessary to support decision-making by the doctor in choosing the optimal patient pharmacotherapy in real time.

Keywords: Information technologies, clinical and pharmaceutical management, newborn, rotavirus infection

COMPARATIVE ASSESSMENT OF BACTERIAL EXCRETION DEGREE DEPENDING FROM SENSIVITY PROFILE OF MYCOBACTERIUM TUBERCULOSIS TO ANTITUBERCULOSIS DRUGS 27-29 Schevchenko O.S., Hovardovska O.O.

One of the important part of the epidemiological component of tuberculosis pathomorphosis in modern conditions is the change in important biological properties of the pathogen, not only in the form of increased frequency and extension of Mycobacterium tuberculosis resistance to anti-tuberculosis drugs, but also changes in manifestations of vital activity - virulence, massivity and growth rate of culture, etc. **The study purpose** was to determine the growth intensity of Mycobacterium tuberculosis depending on resistance to anti-tuberculosis drugs, to assess the bacterial excretions, to compare the data of the phenotypic (Löwenstein-Jensen media) and genotypic method (Xpert MTB/RIF), to carry out the correlation analysis of the results. **Materials and methods:** The results of the phenotypic and genotypic tests of 148 samples have been analyzed. Group 1 included 52 samples with susceptibility of Mycobacterium tuberculosis to anti-tuberculosis drugs, group 2 - 38 samples with monoresistance to isoniazid, group 3 - 58 samples with multi-drug resistance. The bacterial excretion was assessed as: scanty - for single colonies, moderate - for 20 to 100 colonies, massive - for 100 and over. The standard assessment of positive genotypic testing by quantities of DNA copies: very small, small, moderate, great quantity. Correlation analysis has been made in SPSS static program. **Results:** In the 1 group results the following bacterial excretions have been noted: 30,7%- scanty, 28,8%-moderate, 40,3% massive. In the 2 group scanty bacterial excretion was identified in 36,8%, moderate in 39,1%, massive in 23,6%. In the 3 group scanty in 37,9%, moderate in 22,4% and massive bacterial excretion in 39,6%. In the genotypic testing 15 negative results have been noted, which comprised 7,7% in 1 group, 15,8%, in 2 group, 8,6% in 3 group. Very small quantity in 6,2%, 15,6%, 11,3%, Small in 27,1%, 28,1%, 28,3%, Medium quantity in 42,1%, 34,4%, 26,4%, Great quantity in 31,2%, 21,5%, 32,1% - in 1, 2, 3 group, respectively. **Conclusions:** Samples with different resistant strains demonstrated differs in phenotypic and genotypic features. Phenotypic method gave the possibility to detect more than 10% cases with bacterial excretion, that had negative genotypic test. Correlation analysis showed a strong and significant correlation between the indicated results, the Spirmen correlation is 0.75, Kendall - 0.66, with the reliability level $p < 0.01$. Inclusion of the results of genotypic test with information on the quantity of Mycobacterium tuberculosis genetic material in a patient's sputum sample, and also on Mycobacteria tuberculosis resistance, into the assessment of the tuberculosis infection hotbed will enable improvement of epidemiological control.

Keywords: Mycobacterium tuberculosis, tuberculosis, MDR tuberculosis, Xpert MTB/RIF, bacterial excretion, infection control.

COMPARATIVE ANALYSIS OF FREON EXTRACTS FROM LIME FLOWERS CROPPED IN 2016 AND 2017 YEARS 30-35

Demyanenko D. V., Sichkar A. A., Karpenko L. A.

Introduction. The article is devoted to gas-liquid-chromatography analysis of the freon extracts taken from lime flowers cropped in 2016 and 2017 years. It's well-known that quantitative content of a sum of extractives or active substances (and even their structure) can vary considerably in plants depending on harvesting place (and, hence, soils), harvest time, and, especially, weather conditions. This is the main reasons for difficulties in standardization of herbal drugs. Often they are standardized by total extractives, for some plant species there are no techniques of quantitative analysis at all in pharmacopoeias, such an example is lime flowers. Recently, in 2007, 2008, 2009 we studied lipophilic fractions from lime flowers and found slight differences in their quantitative and qualitative compositions. The main aim was to compare chemical composition of volatile compounds in lipophilic freon extracts from lime flowers considering that springs and beginning of summers in 2016 and 2017 was characterized by abnormal weather conditions. **Materials and methods.** The herbal raw materials were lime flowers cropped from linden *Tilia cordata* within Kharkiv region in 2016 and 2017, comminuted to particle size 0,5-2,0 mm. They were extracted with condensed difluorochloromethane (freon R22) in two parallel connected percolators of the experimental equipment. The extraction was two-stage at raw material : extracting solvent ratio 1:8 on each stage under constant circulation of a liquid phase. The minimum pressure and temperature were 12 atm and +30°C accordingly, maximum ones - 17 atm and +45°C. Total exposure of the process took about 2 hours on each stage. The extracts were dissolved in methylene chloride to make 1,00 % solution which was entered into the front injector of gas-chromato-mass-spectrograph «Agilent Technology 6890» (USA) including two consequently connected columns: Agilent 19091S-433HP-5MS, filled with adsorbed 5 % Phenyl-Methyl-Silox, and Agilent 165-6626 USB369911A, and also mass selective detector 5973N. The oven was programmed as follows: equilibration time - 0,5 min, temperature - from 50°C to 250 °C with rate 4°C/min, then 30°C/min to 300 °C, maximum temperature was 325°C, runtime - 51,667 min. Carrier gas was helium: in the first column the flow rate - 1 ml/min under 14,313 psi, and in the second one - 3,8 ml/min at initial pressure 3,8 psi, and in the outlet vacuum was provided. Transferring from gas-liquid chromatograph (GLC) to mass spectrometer occurred at 230°C. The detector and evaporator temperature was supported at 150°C. Electron ionization was spent at 70 eV in range of masses from m/z 29 to 450. Identification of compounds was carried out by comparison of the acquired mass spectra with the data of existing libraries (about 500000 substances). Pentadecane was used as the internal standard. Quantitative content of each substance expressed in percentage of total assay weight of the sample was determined by areas of corresponding peaks considering that 10⁹ standard units of an area corresponded to 0,002 mg of a substance in the assay. Besides, relative percentage content of each substance in the assays was calculated. All solvents and chemicals were of analytical grade. Chromatographic experiments were reproduced in triplicate. **Results and discussion.** On the chromatograms of the difluorochloromethane extracts from lime flowers cropped in 2016 and 2017 on the territory of Kharkiv region 80 and 68 volatile substances were found accordingly. Most of them have retention times more than 35 minutes. Samples of 2017 contained more volatile substances with shorter retention times if compare with extracts obtained from 2016 crop. Total yield of the freon extract in 2016 was 2,75 %, and in 2017 - 1,42 % recalculated on dry weight of raw materials studied. Prevailing chemical groups of substances were aliphatic hydrocarbons and fatty acids, also there were alcohols and

terpenoids in appreciable quantities. In the sample of 2016 total quantitative contents of aliphatic hydrocarbons, aromatic alcohols and terpenoids were approximately 3 times more in comparison with 2017. At the same time, fatty acids, especially unsaturated, and also aldehydes and ketones were found to be much less, and esters were absent at all in 2016 samples. The freon extract of lime flowers cropped in 2016 consisted of 14 substances which relative content exceeded 1 % with total contain 78,7 % of the whole assay, where the dominating ones were: hexatriacontane - 18,73 %, n-hexadecanoic acid - 12,75 %, linoleic alcohol - 10,68 %, 1-pentadecene - 10,57 %. The freon extract of the raw material of 2017 crop included 13 substances with total contain 84,14 % of the whole assay weight from which the prevailing ones are: oleic, octadecanoic, hexadecanoic acids with concentrations 27,68 %, 15,02 % and 12,91 % respectively. **Conclusions.** The comparative analysis of the freon extracts from lime flowers cropped in 2016 and 2017 years using method of gas-liquid chromatography has been carried out. Basing on results of gas-liquid chromatography it has been established that the samples obtained from raw materials of 2016 and 2017 years considerably differ in their quantitative and qualitative characteristics of essential oil fractions: the studied samples contained 80 and 68 compounds accordingly, total percentage of dominating substances was 78,66% and 84,14% of the assays weight respectively. Thus, weather conditions which can change dramatically in spring from year to year in Ukraine, especially in its Northeast part, strongly influence accumulation of volatile compounds in lime flower.

Keywords: difluorochromethane, extraction, lime flower, gas-liquid chromatography.

STUDY OF ANTIMICROBIAL ACTIVITY OF "FUZIPAN-DERMA" GEL

36-38

Bayva P. P., Baranova I. I., Streletz O. P., Makarova O. E.

Introduction. Providing the population with effective drugs for the treatment of infectious diseases of the skin has an important medical and socio-economic significance. Antibiotic resistance of pathogenic microorganisms can contribute to delaying the pathological process, which is an acute problem not only in dermatology, but also in other spheres of medicine. For the comprehensive treatment of infectious diseases of the skin prescribed medicines for local use. Fusidic acid, which is included in the experimental samples of the gel as an active ingredient, is a natural antibiotic that detects a narrow spectrum of antimicrobial activity. Fusidic acid is primarily active in staphylococcus, including those that are methicillin-resistant. Thus, the use of gel with fusidic acid, which exhibits a pronounced antimicrobial effect on the cells of purulent skin inflammations in acne [1, 2, 3] and used to treat complicated staphylococcus infections also [4, 5]. **Materials and methods.** Empirical, theoretical and experimental methods, analysis of professional scientific publications, antimicrobial researches and analysis of statistical data have been used. The subject of the study was to determine the antimicrobial activity of 4 experimental samples of the drug "Fuzipan-derma", with different concentrations of the active pharmaceutical ingredient (API), fusidic acid, in its composition [3, 6] and gel-base without fusidic acid as a control. **Results and discussion.** The results of the studies allow to characterize both the antimicrobial activity of the drug and the speed and completeness of the release of antimicrobial substances from the base, since the zones of growth impairment of microorganisms are formed due to the diffusion of these substances into a dense nutrient medium. In the course of research, one-day suspensions of bacterial microorganisms in a physiological solution were used, and a two-day culture of a yeast-like fungus. The microbial load was 10^7 microbial cells in 1 ml of nutrient medium. The investigated samples No.1, No.2, No.3 does not show fungicidal activity against yeast-like fungus *Candida albicans* ATSC 885-653. Sample No.4 (base of gel) does not show antimicrobial activity in relation to the used cultures of bacteria as well as to the fungus of the genus *Candida albicans*. Samples (No.1, No.2, No.3) exhibit high antimicrobial activity (the diameter of growth impairment zones is 16-25 mm) to gram-positive bacterial cultures *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Streptococcus pyogenes*, *Bacillus subtilis*. It should be noted that the most susceptible to the action of the samples containing fusidic acid is the culture of *Staphylococcus aureus* ATCC 25293. With the increase in the concentration of API (fusidic acid) in the gel, antibacterial activity increases: in relation to *Staphylococcus aureus* ATCC 25293, the diameters of growth impairment zones of the culture are: 25.6 ± 0.5 mm (sample number 1), 33.8 ± 0.4 mm (sample number 2), 35.4 ± 0.5 mm (sample number 3); to the culture of *Staphylococcus epidermidis* ATCC 12228: 23.4 ± 0.5 , 29.6 ± 0.5 , 31.8 ± 0.4 mm respectively 0.5%, 1.0% and 2.0%; to the culture of *Streptococcus pyogenes* ATCC 12344: 24.2 ± 0.4 , 31.6 ± 0.5 , 33.6 ± 0.5 , respectively, samples with a concentration of fusidic acid of 0.5%, 1.0%, and 2.0%. In relation to the gram-positive spore culture of *Bacillus subtilis* ATCC 6633, gel samples showed activity at the level of 20.8 ± 0.4 , 23.6 ± 0.5 , 25.4 ± 0.5 , depending on the content of fusidic acid. The research showed that gel samples with fusidic acid content of 3% had a higher antimicrobial activity than 2.0% gel, but in order to minimize the risk of local allergic reactions and reduce the resorptive action of the antibiotic and the development of microbial resistance, it is advisable to select a sample of fusidic acid of 2.0% as a sufficiently effective API concentration. **Conclusions.** For further studies for development the technology of gel with fusidic acid for the treatment of acne, it is advisable to select sample number 2 with a content of fusidic acid of 2.0%.

Keywords: antimicrobial activity, biological researches, fusidic acid, dermatological medicines