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# ВЕСТНИК ФАРМАЦИИ

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**Для науковців, провізорів, лікарів, організаторів системи охорони здоров'я.**

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# СИНТЕЗ ТА АНАЛІЗ БІОЛОГІЧНО АКТИВНИХ РЕЧОВИН

*Recommended by Doctor of Chemistry, professor M.Ye.Blazheyevskiy*

UDC 543.257+547.583.5

## THE REACTIVITY OF N-[(2-OXOINDOLIN-3-YLIDENE)-2-OXIACETYL] AMINO ACIDS

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*Key words: N-[(2-oxoindolin-3-ylidene)-2-oxiacetyl] amino acids; reactivity; correlation analysis; Hammett equation*

*The reactivity of N-[(2-oxoindolin-3-ylidene)-2-oxiacetyl] amino acids has been investigated in the reversible conditions by studying their acid-base properties in the binary solvent of dioxane – water (60 volume % of dioxane) at 25°C. The experimental compounds have been proven to be weak dibasic acids. Their pKa values have been determined by Noyes method. The correlation of these values to both of the reactive sites (COOH- and OH-groups) has been performed. It has been shown that each $CH_2$- prolongation step of the polymethylene chain reduces acidity of compounds at both reactive sites of ionization. Hammett correlation equations ($pKa_{1,2}$ – f(σ)) have been calculated for N-[(2-oxoindolin-3-ylidene)-2-oxiacetyl] amino acids, and it allows to predict the acid-base properties of compounds of the given homological series. The low sensitivity of the reactive sites towards polymethylene chain prolongation has been found. The results obtained are used for mathematical modeling of QSAR-analysis of the compounds of the isostructural series.*

N-[(2-oxoindolin-3-ylidene)-2-oxiacetyl] amino acids and their derivatives possess various types of the biological activity [6-9]. That is why this class of compounds is intensively used for purposeful search of active pharmacophores. The pharmacological activity depends on the ability of a pharmacophore to form complexes with biological receptors, and it, in turn, is determined by the pharmacophore reactivity, in particular its acid-base properties. Therefore, the study of the reactivity of homological series of these biologically active substances is of great scientific and practical interest in connection with the possibility of optimization of their targeted synthesis and modeling of the active pharmacophores. Data describing the reactivity of oxoindole derivatives are absent in the chemical and biological scientific literature sources.

**Materials and Methods**

The synthetic studies and physico-chemical characteristics of the experimental series of N-[(2-oxoindolin-3-ylidene)-2-oxiacetyl] amino acids were presented in our previous works [3].

The ionization constants of compounds I-VI (see Scheme) were determined by the potentiometric titration method [1]. The titrant was 0.05 M aqueous solution of potassium hydroxide free of $CO_2$. The concentration of the solutions to be titrated was 0.005 M at the point of semineutralization. The measurements were performed on an EV-74 ionomer using two electrodes: a glass (ESP 43-074) indicatory one, as well as a saturated chlorine-silver electrode. The latter was applied as a reference electrode. Determinations were carried out at 25°C in triplicates. The accuracy of the results obtained was estimated by methods of mathematical statistics of small samples (confidence probability – 0.95) [4].

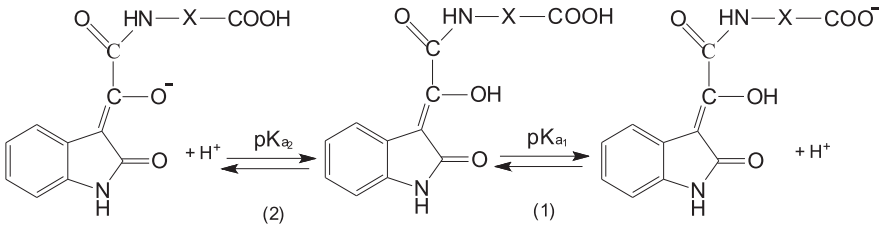
The mixed solvent of dioxane – water (60 volume % of dioxane) was prepared from freshly bi-distilled water free of $CO_2$ and 1,4-dioxane (very pure) without additional purification.

**Results and Discussion**

The reactivity of N-[(2-oxoindolin-3-ylidene)-2-oxiacetyl] amino acids was studied in the reversible conditions on the model of the acid-base equilibria (Scheme).

Ionization constants ($pKa_1$ and $pKa_2$) of the corresponding acid-base equilibria (1) and (2) of the compounds studied are given in Table.

The preliminary analysis of the titration curves of compounds I-VI obtained by the electrometric method has demonstrated that these substances are dibasic acids, which $\Delta pKa(pKa_2 - pKa_1)$ is less than 4. Therefore, the classic Noyes method was chosen for calculation [1]. The pKa ionization values of ethyl esters of the corresponding N-[(2-oxoindolin-3-ylidene)-2-oxiacetyl] amino acids previously obtained and described in our works [2] were thoroughly analyzed to make correlations of the pKa values obtained to the certain reactive sites of acidic ionization. The high pKa values were referred to enolic hydroxyl ionization (equilibrium 2). This fact corresponds to the literature data concerning a higher acidity of the carboxyl group compared to the enolic one [10].



**I**: X = 0; **II**: X = $CH_2$; **III**: X = $(CH_2)_2$; **IV**: X = $(CH_2)_3$; **V**: X = $(CH_2)_4$; **VI**: X = $(CH_2)_5$

Scheme

From the table data it follows that with each $CH_2$-prolongation step of the polymethylene chain length in the amino acid molecular fragment the ionization degree of both reactive sites of the molecules in the compounds studied decreases. However, the degree of the isolating effect of methylene groups on the reactive sites is different:

1. acidity alteration of COOH-site: $\Delta pKa = pKa_6 - pKa_1 = -0.26$;

2. acidity alteration of OH-site: $\Delta pKa = pKa_6 - pKa_1 = -0.29$.

This fact indicates that sensitivity of the carboxylic group to the induction effect transmission is higher than that of the enolic hydroxyl.

The $\Delta pKa$ value ($\Delta pKa = pKa_{n+1} - pKa_n$; where *n* is the number of $CH_2$ – groups) of both reactive sites remains constant almost everywhere (see Table). This allowed us to perform the qualitative evaluation of dependence of pKa values of both reactive sites on the length (n) of the polymethylene molecular fragment using the principle of free energies linearity by the correlation analysis method.

The COOH-reactive site:

$$pKa_1 = (5.34\pm0.06) + (0.052\pm0.002)\cdot n \qquad (1)$$
$$n=6 \quad s=0.003 \quad r=0.9970$$

The OH-reactive site:

$$pKa_2 = (6.43\pm0.05) + (0.039\pm0.002)\cdot n \qquad (2)$$
$$n=6 \quad s=0.002 \quad r=0.9950$$

The electron effect of substituents was estimated by Hammett equation ($pKa = a + \rho\cdot\sigma$). The $CH_2$-fragment constant ($\sigma$) was chosen as 0.388 according to the classic work of Palm V.A. [5]. The $CH_2$-fragment constants were calculated by the following formula:

$$\sigma(CH_2)_n = 0.388\cdot n$$

The equations of $pKa - f(\sigma)$ correlation for both reactive sites with statistically significant parameters were obtained.

The COOH-reactive site:

$$pKa_1 = (5.35\pm0.01) + (0.133\pm0.005)\cdot\sigma \qquad (3)$$
$$n=6 \quad s=0.002 \quad r=0.9970$$

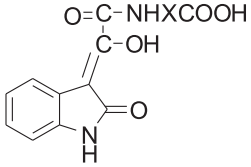
The OH-reactive site:

$$pKa_2 = (6.43\pm0.01) + (0.105\pm0.006)\cdot\sigma \qquad (4)$$
$$n=6 \quad s=0.003 \quad r=0.9950$$

The parameters of equations (3,4) indicate weakening of the ionization degree at both reactive sites with each $CH_2$- prolongation step of the polymethylene chain length, i.e. the isolating effect of methylene groups [5], since the reaction constants $\rho_1$ and $\rho_2$ are positive. However, $\rho_1 > \rho_2$, and it indicates a somewhat higher sensitivity of the carboxylic group compared to the enolic one towards the isolating effect of the polymethylene chain. It should be noted that $\rho_1$ and $\rho_2$ values are extremely

Table

Acid-base equilibria of N-[(2-oxoindolin-3-ylidene)-2-oxiacetyl] amino acids



| Compound | $pKa_1$ | $\Delta pKa_1 = pKa_{n+1} - pKa_n$ | $pKa_2$ | $\Delta pKa_2 = pKa_{n+1} - pKa_n$ |
|---|---|---|---|---|
| I | 5.35±0.03 | – | 6.44±0.03 | – |
| II | 5.41±0.05 | 0.06 | 6.47±0.04 | 0.03 |
| III | 5.44±0.05 | 0.03 | 6.51±0.05 | 0.04 |
| IV | 5.50±0.03 | 0.06 | 6.54±0.02 | 0.03 |
| V | 5.56±0.04 | 0.06 | 6.60±0.04 | 0.06 |
| VI | 5.61±0.03 | 0.05 | 6.63±0.05 | 0.03 |

low, i.e. attenuation of the electron effects transmission was yet insignificant.

CONCLUSIONS

1. The reactivity of N-[(2-oxoindolin-3-ylidene)-2-oxiacetyl] amino acids has been investigated by studying their acid-base equilibria in the reversible conditions.

2. The experimental compounds have been proven to be weak dibasic acids. Their ionization process equations have been suggested.

3. The ionization constants values ($pKa_2$ and $pKa_1$) of six N-[(2-oxoindolin-3-ylidene)-2-oxiacetyl] amino acids have been determined. It has been shown that prolongation of the polymethylene chain reduces ionization at both reactive sites (COOH and OH).

4. The influence of methylene links in the amino acid molecular fragment has been quantitatively estimated according to Hammett equation, and the low sensitivity of both reactive sites towards the polymethylene chain prolongation has been found.

REFERENCES


1. *Альберт А., Сержент Е. Константы ионизации кислот и оснований. – М.: Химия, 1964. – 214 с.*
2. *Колесник С.В., Свечникова Е.Н., Святская Т.Н. и др. // Научные ведомости Белгородского государственного университета. – 2014. – Вып. 25, №4(175). – С. 240-243.*
3. *Колісник С.В., Болотов В.В., Ляшенко О.В. // ЖОФХ. – 2009. – Т. 7, вип. 4 (28). – С. 55-59.*
4. *Львовский Е.Н. Статистические методы построения эмпирических формул. – М.: Высш. шк., 1988. – 125 с.*
5. *Пальм В.А. Основы количественной теории органических соединений. – Л.: Химия, 1967. – 356 с.*
6. *Пат. на винахід 85318 (2009) Україна, МПК C 07 C 229/02 (2006.01), A 61 K 31/197 (2006.01). Етиловий ефір N-(2-оксоіндолін-3-гліоксилоїл)-ε-амінокапронової кислоти, який проявляє діуретичну, антигіпоксичну та протизапальну дію / В.В.Болотов, С.В.Колісник, А.І.Березнякова, І.І.Шевцов. – Заявл.: 23.07.07. Опубл.: 12.01.09. – Бюл. №1.*
7. *Пат. на винахід 91166 (2010) Україна, МПК A 61 K 31/404 (2006.01), A 61 K 31/405 (2006.01), C 07 D 209/34 (2006.01), A 61 P 25/28 (2006.01), A 61 P 25/22 (2006.01). Застосування етилового естеру N-[(2-оксоіндолініліден-3)-2-оксіацетил]-амінооцтової кислоти як ноотропного та анксіолітичного засобу / В.В.Болотов, С.В.Колісник, С.Ю.Штриголь, О.В.Шатілов. – Заявл.: 17.08.09. Опубл.: 25.06.10. – Бюл. №12.*
8. *Пат. на винахід 95533 (2011) Україна, МПК C 07 D 209/34 (2006.01), A 61 K 31/404 (2006.01), A 61 P 25/00 (2006.01). Похідні N-[(2-оксоіндолініліден-3)-2-оксіацетил]-4-амінобутанової кислоти, які проявляють ноотропну дію / В.В.Болотов, С.В.Колісник, С.Ю.Штриголь, О.В.Шатілов. – Заявл.: 25.12.09. Опубл.: 10.08.11. – Бюл. №15.*
9. *Пат. на винахід 99014 (2012) Україна, МПК C 07 C 227/14 (2006.01), A 61 K 31/191 (2006.01), A 61 K 31/195 (2006.01), A 61 P 29/00 (2006.01). Метиловий естер N-[(2-оксоіндолініліден-3)-2-оксіацетил]-амінооцтової кислоти, який проявляє протизапальну дію / В.В.Болотов, С.В.Колісник, Т.І.Тюпка, Т.О.Лой, А.В.Міщенко. – Заявл.: 10.11.10. Опубл.: 10.07.12. – Бюл. №13.*
10. *Черных В.П., Бризицкая О.А., Свечникова Е.Н. и др. // ЖОХ. – 2004. – Т. 74, вып. 4. – С. 674-677.*


**РЕАКЦІЙНА ЗДАТНІСТЬ N-[(2-ОКСОІНДОЛІН-3-ІЛІДЕН)-2-ОКСІАЦЕТИЛ]АМІНОКИСЛОТ**

***С.В.Колісник, О.М.Свєчнікова, Р.И.Кратенко, О.В.Колісник***

***Ключові слова:*** *N-[(2-оксоіндолін-3-іліден)-2-оксіацетил]амінокислоти; реакційна здатність; кореляційний аналіз; рівняння Гамета*

*Досліджена реакційна здатність N-[(2-оксоіндолін-3-іліден)-2-оксіацетил]амінокислот в обернених умовах шляхом вивчення їх кислотно-основних рівноваг у бінарному розчиннику діоксан-вода (60 об'ємних % діоксану) при 25°С. Показано, що ці сполуки – слабкі двоосновні кислоти. Їх рКа визначали за методом Нойєса. Проведено співвіднесення цих значень з двома реакційними центрами (СООН та ОН-групи). Показано, що подовження поліметиленового ланцюга зменшує кислотність сполук за обома центрами іонізації. Розраховані кореляційні рівняння Гамета $pKa_{1,2}$ – f(σ) для N-[(2-оксоіндолін-3-іліден)-2-оксіацетил]амінокислот, що дозволяє прогнозувати кислотно-основні властивості сполук цього гомологічного ряду. Встановлена низька чутливість реакційних центрів до подовження поліметиленового ланцюга. Одержані результати використовуються для математичного моделювання QSAR – аналізу сполук цього ізоструктурного ряду.*

**РЕАКЦИОННАЯ СПОСОБНОСТЬ N-[(2-ОКСОИНДОЛИН-3-ИЛИДЕН)-2-ОКСИАЦЕТИЛ] АМИНОКИСЛОТ**

***С.В.Колесник, Е.Н.Свечникова, Р.И.Кратенко, Е.В.Колесник***

***Ключевые слова:*** *N-[(2-оксоиндолин-3-илиден)-2-оксиацетил] аминокислоты; реакционная способность; корреляционный анализ; уравнение Гаммета*

*Исследована реакционная способность N-[(2-оксоиндолин-3-илиден)-2-оксиацетил]аминокислот в обратимых условиях путем изучения их кислотно-основных свойств в бинарном рас-*

*творителе диоксан-вода (60 объемных % диоксана) при 25°С. Показано, что эти соединения – слабые двухосновные кислоты. Их рКа определялись по методу Нойеса. Проведено соотнесение этих значений с двумя реакционными центрами (СООН и ОН-группы). Показано, что удлинение полиметиленовой цепи уменьшает кислотность соединений по обоим центрам ионизации. Рассчитаны корреляционные уравнения Гаммета $pKa_{1,2} – f(\sigma)$ для N-[(2-оксоиндолин-3-илиден)-2-оксиацетил]аминокислот, что позволяет прогнозировать кислотно-основные свойства соединений этого гомологического ряда. Установлена низкая чувствительность реакционных центров к удлинению полиметиленовой цепи. Полученные результаты используются для математического моделирования QSAR – анализа соединений этого изоструктурного ряда.*

*Recommended by Doctor of Pharmacy, professor S.V.Kolisnyk*

UDC 547.398.1:547.461.3

# PHYSICOCHEMICAL PARAMETERS AND THE DIURETIC ACTIVITY OF 5-(4-R)BEHZYL-1,3,4-OXADIAZOL-2-IL-THIOACETIC ACID AMIDES

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*Key words: 1,3,4-oxadiazol; "drug likeness" concept; Lipinski's Rules of Five; diuretic activity; cytotoxicity*

*The group of aryl(heteryl)amides of 5-(4-R)benzyl-1,3,4-oxadiazol-2-il- thioacetic acid has been tested for compliance with the "drug likeness" concept. A number of physical and chemical properties that determine bioavailability according to Lipinski's Rule of Five has been calculated using the ACD/Labs computer programme. It has been found that these compounds can be recommended for further research as compounds with favourable physicochemical properties according to Lipinski's Rule of Five. The results of the experimental study of the ability of new substances to stimulate the urinary renal function predicted by the computer PASS programme have shown that some of the new compounds synthesized are promising diuretics. The results of studying cytotoxicity in vitro for the most promising compounds have shown that these compounds possess low toxicity. The pharmacological screening has allowed to select two promising compounds that are relevant for further study.*

There are many approaches that assess a compound's "*drug likeness*", partially based on topological descriptors or other properties. In addition, the biological activity is also usually a function of the complex influence of a number of molecular descriptors. Lipinski's rule of Five also known as the Rule of Five is a rule of thumb to evaluate drug likeness or determine if a chemical compound with a certain pharmacological or biological activity has properties that would make it a likely orally active drug in human. The rule was formulated by Christopher A.Lipinski in 1997. It is based on the observation that active substances of most drugs are relatively small and lipophilic molecules [16]. The rule describes molecular properties that are important for drug pharmacokinetics in the human body, including their absorption, distribution, metabolism, and excretion ("ADME"). However, the rule does not predict if a compound is pharmacologically active.

Among heterocyclic compounds, 1,3,4-oxadiazole has become an important construction scaffold for development of new drugs. Compounds containing 1,3,4-oxadiazole ring have a broad spectrum of the biological activity, including the anticancer [11], anticonvulsant [17], anti-inflammatory [14], antiviral [8], antifungal [12], antimycobacterial action [13]. The oxadiazole ring occurs frequently in a variety of pharmaceutical drugs, including Furamizol, Fenadiazole, Zibotentan, Picovir, Raltegravir, Butalamine, Fasiplon and Oxolamine [10].

The results of computer prognosis of the biological activity spectrum of amides 5-(4-R)benzyl-1,3,4-oxadiazol-2-il-thioacetic acid have shown the possible diuretic activity (activity indexes of this compounds are in the range of 0.496 to 0.595) [15]. Expediency of searching potential diuretics among derivatives of 1,3,4-oxadiazoles is also confirmed by the literature data [10].

The aim of this work was to test amides of 5-(4-R) benzyl-1,3,4-oxadiazol-2-il-thioacetic acid for compliance with the concept of "drug likeness' and study their diuretic activity and toxicity.

**Materials and Methods**

The synthetic methods used to prepare amides of 5-(4-R)benzyl-1,3,4-oxadiazol-2-il-thioacetic acid can be found in previously reported works [1-2], characteristics of the substances synthesized are given in Tab. 1.

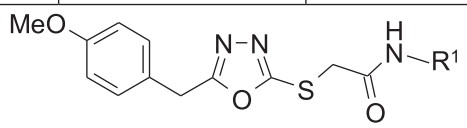
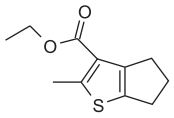
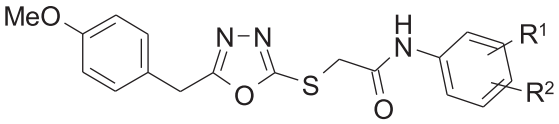
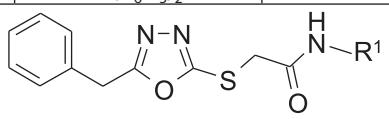
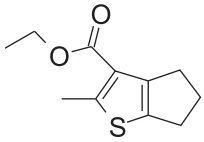
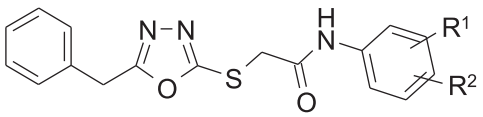
A number of physical and chemical properties determining bioavailability for aryl (heteryl)amides of 5-(4-R)benzyl-1,3,4-oxadiazol-2-il-thioacetic acid ***1.1-1.20*** was calculated using the ACD/Labs computer programme. The purpose of calculations was to compare the values obtained with the desired values of descriptors according to the concept of "drug likeness" followed by elimination of unwanted molecules from databases to optimize further screening. The results obtained are presented in Tab. 2.

The ability of the substances synthesized stimulate or, conversely, inhibit the urinary renal function was studied by the method of E.B. Berkhin in nonlinear white male rats weighing 200.0-250.0 g [4]. Hypothiazide in the dose of 40 mg/kg. was selected as the reference drug when determining the diuretic activity. The compounds studied and the reference drug were introduced as a single intragastrical injection in the form of a finely dispersed aqueous suspension stabilized by Tween-80.

All laboratory animals were received a water test in the amount of 3% of the body weight. After introduction of the substances rats were placed in the "exchange

Table 1

Characteristics of 5-(4-R)benzyl-1,3,4-oxadiazol-2-il-thioacetic acid ***1.1-1.20*** amides

| Compound | $R^1$ | $R^2$ |
|---|---|---|
| |  | |
| 1.1 |  | – |
| |  | |
| 1.2 | H | H |
| 1.3 | 4-$C_2H_5$ | H |
| 1.4 | 4-$COOC_2H_5$ | H |
| 1.5 | 2-$CH_3$ | 3-$CH_3$ |
| 1.6 | 3-Cl | 4-Cl |
| 1.7 | 2-Cl | 5-$CF_3$ |
| 1.8 | 2-$CH_3$ | 3-Cl |
| 1.9 | 4-$OC_2H_5$ | H |
| 1.10 | 3-$CH_3$ | H |
| 1.11 | naphtyl | - |
| 1.12 | $N(C_6H_5)_2$ | - |
| |  | |
| 1.13 |  | |
| |  | |
| 1.14 | H | H |
| 1.15 | 4-$C_2H_5$ | H |
| 1.16 | 4-$COOC_2H_5$ | H |
| 1.17 | 2-$CH_3$ | 3-$CH_3$ |
| 1.18 | 4-$CH_3$ | H |
| 1.19 | 3-Cl | 4-Cl |
| 1.20 | 2-Cl | 5-$CH_3$ |

cages". The amount of urine excreted by the control group of animals was taken as 100%. The control group of rats received water and Tween-80 in the same volume. The results obtained were compared with the data of the control group of animals. Indicators of the diuretic activity were the total amount of urine within 4 hours and the amount of urine per 100 g of the animal's body weight.

Recently, the so-called alternative methods are increasingly used in advanced preclinical trials along with the traditional methods. Today, alternative methods include the use of invertebrate organisms, plants, microorganisms, cell cultures, as well as a number of physical and biological methods. Using biological models (*in vitro*) can explain biological phenomena, which are difficult to investigate in the experiments with animals because of the complexity of interaction between different effectors and inhibitors.

The main preconditions for wider implementation in practice of preclinical studies of alternative methods, including methods of cell cultures – model test systems, are ethical considerations about exclusion or limitation of experiments on warm-blooded animals [3]. It should be noted that introduction of *in vitro* methods provides pharmacological and toxicological screening systems for development of new substances, allows to use biological systems derived from genetically modified animals, their certain metabolic components are close to human, and create a variety of conditions simultaneously, i.e. accelerate and increase the reliability of research, development and introduction of new drugs [5].

The study of toxicity of the most active compounds *in vitro* was conducted on the model test systems of the bone marrow cells of rats. The compounds were added to the cell suspension in the ratio of 1: 1 in the dose of 15 mg/ml. The cell activity in all samples was measured in 30 min after the start of the experiment. Counting live and dead cells was carried out with the Goryaev chamber after staining with trypan blue [7]. The proportion of dead cells was determined by the formula:

$$Pr = \frac{Nm}{N\ total} \cdot 100\%,$$

where: Nm – is the number of dead cells in the sample; N total – is the total number of cells in the sample.

Further the experimental results were statistically processed using a "StatSoft" software package [6] (Tab. 4).

**Results and Discussion**

All compounds studied by the complex of physical and chemical properties meet modern requirements for new compounds when testing their biological activity. The results obtained have shown that all substances can be recommended for further research as compounds with favourable physicochemical properties.

The diuretic activity of aryl (heteryl)amides of 5-(4-R)-benzyl-1,3,4-oxadiazol-2-il-thioacetic acid ***1.1-1.20*** are shown in Table 3. In terms of the total amount of urine substances ***1.3*** and ***1.4*** are the most promising. By the diuretic activity **(**the difference in % relative to the control) ***1.17*** and ***1.20*** are the most promising.

Comparing the experimental data obtained (Tab. 3) it can be concluded that substances ***1.17*** and ***1.20*** are the most active in comparison with the control.

The results of the study of cytotoxicity (Tab. 4) have shown that the compounds synthesized have low toxicity (Pr<20) and allowed to choose two substances (***1.17, 1.20)***, which are promising for further study.

Table 2

Determination of parameters of 5-(4-R)benzyl-1,3,4-oxadiazol-2-il-thioacetic acid amides ***1.1-20*** according to Lipinski's Rule of Five

| Compound | Parameters of compounds | | | | |
|---|---|---|---|---|---|
| | Molecular weight, MW | Molar refraction, MR cm | Partition coefficient, Log P | Number of the hydrogen bond donors | Number of the hydrogen bond acceptors |
| 1.1 | 268.24 | 67.14 | 2.6 | 5 | 3 |
| 1.2 | 252.28 | 71.57 | 2.3 | 3 | 3 |
| 1.3 | 383.46 | 105.54 | 2.5 | 5 | 3 |
| 1.4 | 427.47 | 112.03 | 2.9 | 5 | 3 |
| 1.5 | 383.46 | 105.53 | 2.7 | 3 | 3 |
| 1.6 | 424.30 | 105.94 | 2.4 | 5 | 3 |
| 1.7 | 457.85 | 106.10 | 2.3 | 3 | 3 |
| 1.8 | 403.88 | 105.73 | 2.5 | 2 | 3 |
| 1.9 | 369.43 | 105.98 | 2.8 | 3 | 3 |
| 1.10 | 369.43 | 100.91 | 2.9 | 3 | 3 |
| 1.11 | 405.46 | 113.84 | 2.4 | 3 | 3 |
| 1.12 | 431.50 | 121.22 | 2.5 | 3 | 3 |
| 1.13 | 443.53 | 115.64 | 2.6 | 4 | 3 |
| 1.14 | 325.38 | 89.92 | 2.4 | 2 | 3 |
| 1.15 | 369.43 | 100.91 | 2.9 | 4 | 3 |
| 1.16 | 397.44 | 105.66 | 2.8 | 4 | 3 |
| 1.17 | 353.43 | 99.16 | 2.7 | 2 | 3 |
| 1.18 | 339.41 | 94.54 | 2.4 | 2 | 3 |
| 1.19 | 394.27 | 99.57 | 2.4 | 4 | 3 |
| 1.20 | 427.82 | 99.73 | 2.7 | 2 | 3 |
| The range of values | 252.28-457.85 | 67.14-121.22 | 2.3-2.9 | 2-5 | 3 |
| The average value | 355.07 | 94.18 | 2.6 | 3.5 | 3 |
| The maximum permissible value | 460 | 130 | 5.6 | 5 | 10 |
| The optimal value | 357 | 97 | 2.52 | – | – |

Table 3

The diuretic activity of 5-R-benzyl-1,3,4-oxadiazol-2-il-thioacetic acid amides

| Compound | The average dose of the substance, mg/kg | The average weight of the rat, g | The average total amount of urine, ml | The amount of urine per 100 g of the animal's weight, ml | Diuretic activity, % | The difference in % relative to the control |
|---|---|---|---|---|---|---|
| 1 | 2 | 3 | 4 | 5 | 6 | 7 |
| 1.1 | 34.4 | 213.33 | 3.87 | 1.81 | 58.22 | -41.78 |
| 1.2 | 33.04 | 208.33 | 6.27 | 3.00 | 96.44 | -3.56 |
| 1.3 | 31.84 | 235.00 | 7.33 | 3.10 | 99.66 | -0.34 |
| 1.4 | 29.74 | 241.67 | 7.73 | 3.20 | 102.93 | 2.93 |
| 1.5 | 28.34 | 218.33 | 5.67 | 2.57 | 82.59 | -17.41 |
| 1.6 | 29.84 | 243.33 | 6.77 | 2.80 | 117.09 | 17.09 |
| 1.7 | 28.43 | 230.00 | 6.40 | 2.78 | 116.43 | 16.43 |
| 1.8 | 28.13 | 230.00 | 5.13 | 2.23 | 93.39 | -6.61 |
| 1.9 | 27.23 | 240.00 | 6.37 | 2.66 | 111.25 | 11.25 |
| 1.10 | 29.74 | 226.67 | 6.83 | 3.01 | 125.93 | 25.93 |
| 1.11 | 28.34 | 226.67 | 6.53 | 2.89 | 120.85 | 20.85 |
| 1.12 | 28.64 | 206.67 | 5.30 | 2.57 | 128.90 | 28.90 |
| 1.13 | 31.24 | 206.67 | 5.27 | 2.42 | 121.53 | 21.53 |

*Table 3 continued*

| 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---|---|---|---|---|---|---|
| 1.14 | 31.24 | 220.00 | 4.20 | 1.91 | 95.83 | -4.17 |
| 1.15 | 34.04 | 220.00 | 4.20 | 1.91 | 129.85 | 29.85 |
| 1.16 | 40.25 | 211.67 | 5.47 | 2.58 | 137.52 | 37.52 |
| 1.17 | 29.84 | 246.67 | 5.50 | 2.26 | 157.21 | 57.21 |
| 1.18 | 29.84 | 228.33 | 5.87 | 2.58 | 131.47 | 31.47 |
| 1.19 | 32.64 | 228.33 | 4.97 | 2.16 | 120.22 | 20.22 |
| 1.20 | 38.85 | 233.33 | 6.17 | 2.65 | 161.48 | 61.48 |

Table 4

Assessment of the death rate of bone marrow cells in rats

| Compound | Pr, % |
|---|---|
| Control | 5.23±0.23 |
| 1.3 | 19.94±0.42 |
| 1.4 | 19.27±0.47 |
| 1.17 | 7.91±0.08 |
| 1.20 | 10.03±0.65 |

## CONCLUSIONS

1. Testing for compliance with the concept of "drug likeness" of 20 structures of 5(4-R)benzyl-1,3,4-oxadiazol-2-il-thioacetic acid derivatives has been performed. The results obtained have shown that the compounds obtained can be recommended for further study as compounds with favourable physicochemical properties according to Lipinski's Rule.

2. The study of the diuretic activity and toxicity of the compounds synthesized (*in vitro*) has identified two active compounds, which are relevant for further study.

## REFERENCES


1. *Георгіянц В.А., Перехода Л.О., Рибальченко Т.Л. та ін. // Фармац. журн. – 2010. – №6. – С. 25-31.*
2. *Георгіянц В.А., Перехода Л.О., Рядних К.С. та ін. // Тези доп. наук. конф. «Фармація України. Погляд у майбутнє: матер. VII Нац. з'їзду фармац. України». – Х., 2010. – Т. 1. – С. 86.*
3. *Доброва В.Е., Должикова Л.М., Малоштан Е.А. // Тези докл. научн. конф. «Фармация Казахстана: интеграция науки, образования и производства». – К., 2009. – С. 45-49.*
4. *Доклінічні дослідження лікарських засобів: Метод. рекоменд. / Под ред. А.В.Стефанова. – К., 2001. – С. 371-395.*
5. *Коваленко В.М., Стефанов О.В., Максимов І.М. Методичні рекомендації по експериментальному вивченню токсичної дії лікарських засобів. – К.: МОЗ України, Державний фармакологічний центр, 2000. – С. 43.*
6. *Лапач С.Н., Чубенко А.В., Бабич П.Н. Статистические методы в медико-биологических исследованиях с использованием Excel. – К.: Морион, 2000. – 320 с.*
7. *Чехун В.Ф., Кулік Г.І., Шарикіна Н.І. Методичні рекомендації з вивчення канцерогенних властивостей нових речовин та лікарських засобів. – К.: МОЗ України, Державний фармакологічний центр, 2001. – С. 19.*
8. *Farghaly A.R., El-Kashef H. // Arkivoc. – 2006. – Vol. 11. – P. 76-90.*
9. *Kalpesh P., Jayachandran E., Ravi S. et al. // Intern. J. of Pharma and Bio Sci. – 2010. – Vol. 1, №3. – P. 125-135.*
10. *Khader A.M. // Der Pharma Chemica. – 2013. – Vol. 5 (2). – P. 24-32.*
11. *Kumar A., D'Souza S.S., Gaonkar S.L. et al. // Invest. New Drugs. – 2008. – Vol. 26. – P. 425-435.*
12. *Liu F., Luo X., Song B. et al. // Bioorg. Med. Chem. – 2008. – Vol. 16. – P. 3632-3640.*
13. *Macaev F., Rusu G., Pogrebnoi S. et al. // Bioorg. Med. Chem. – 2005. – Vol. 13. – P. 4842-4850.*
14. *Narayana B., Raj K.K., Ashalatha B.V. et al. // Arch. Pharm. – 2005. – Vol. 338. – P. 373-377.*
15. *PASS: Prediction of Activity Spectra for Substances. http:///www.ibmh.msk.su/PASS.*
16. *Veber D.F., Johnson S.R., Cheng H.Y. et al. // J. Med. Chem. – 2002. – Vol. 45, №12. – P. 2615-2623.*
17. *Zarghi A., Tabatabai S.A., Faizi M. et al. // Bioorg. Med. Chem. Lett. – 2005. – Vol. 15. – P. 1863-1865.*

**ФІЗИКО-ХІМІЧНІ ПАРАМЕТРИ ТА ДІУРЕТИЧНА АКТИВНІСТЬ АМІДІВ 5-(4-R) БЕНЗИЛ-1,3,4-ОКСАДІАЗОЛ-2-ІЛ-ТІОАЦЕТАТНОЇ КИСЛОТИ**

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***Ключові слова:*** *1,3,4-оксадіазол; концепція «схожість з ліками»; Правила Ліпінські; діуретична активність; цитотоксичність*

*Група арил(гетерил)амідів 5-(4-R)бензил-1,3,4-оксадіазол-2-іл-тіоацетатної кислоти була протестована на відповідність концепції «схожість з ліками», для чого за допомогою комп'ютерної програми ACD/Labs розраховані їх фізико-хімічні параметри, що визначають біодоступність за «Правилами Ліпінські». Встановлено, що зазначені сполуки можуть бути рекомендовані для подальшого вивчення як такі, що згідно з Правилами Ліпінські мають сприятливі фізико-хімічні параметри. Результати вивчення сечогінної активності, що прогнозується для даної групи похідних 1,3,4-оксадіазолу за даними комп'ютерної програми PASS, показали, що деякі досліджені аміди є перспективними діуретиками. Визначенням токсичності (in vitro) цих сполук з'ясовано, що вони мають низьку токсичність. Проведений фармакологічний скринінг дозволив відібрати для поглибленого дослідження дві найбільш активні БАР.*

**ФИЗИКО-ХИМИЧЕСКИЕ ПАРАМЕТРЫ И ДИУРЕТИЧЕСКАЯ АКТИВНОСТЬ АМИДОВ 5-(4-R) БЕНЗИЛ-1,3,4-ОКСАДИАЗОЛ-2-ИЛ-ТИОУКСУСНОЙ КИСЛОТЫ**

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***Ключевые слова:*** *1,3,4-оксадиазол; концепция «сходство з лекарствами»; Правила Липински; диуретическая активность; цитотоксичность*

*Группа арил (гетерил) амидов 5 (4-R) бензил-1,3,4-оксадиазол-2-ил-тиоацетатной кислоты была протестирована на соответствие концепции «сходство с лекарствами», для чего с помощью компьютерной программы ACD/Labs рассчитаны их физико-химические параметры, определяющие биодоступность по «Правилам Липински». Установлено, что указанные соединения могут быть рекомендованы для дальнейшего изучения как такие, которые согласно «Правил Липински» имеют благоприятные физико-химические параметры. Результаты изучения мочегонной активности, которая прогнозируется для данной группы производных 1,3,4-оксадиазола по данным компьютерной программы PASS, показали, что некоторые исследованные амиды являются перспективными диуретиками. Определением токсичности (in vitro) этих соединений установлено, что они имеют низкую токсичность. Проведенный фармакологический скрининг позволил отобрать для углубленного исследования два наиболее активных БАВ.*

*Recommended by Doctor of Pharmacy, professor V.S.Bondar*

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# THE STUDY OF AMINO ACID COMPOSITION OF SAPROPEL BY THE CAPILLARY ELECTROPHORESIS METHOD

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*Key words: sapropel; capillary electrophoresis method; amino acid composition*

*A diverse composition of sapropels allows to use them successfully in cosmetology. Sapropelic therapeutic muds exhibit the anti-inflammatory and desensitizing effect, protect the body from the damaging effects of free radicals, slow down the aging processes, give the skin freshness and elasticity, improve the cell regeneration by 10%, moisturize and increase elasticity and thickness of the keratoid layer of the epidermis. A wide range of indications for using sapropels demonstrates the importance of their application for prevention and treatment of diseases, and the relevance and prospects for their further studies to obtain new pharmaceuticals and cosmetics. When conducting research by the capillary electrophoresis method the "Kapel' – 105/105 M" system with a positive polarity of a high voltage source (the internal diameter of the capillary is 50 μm, the full length of the capillary is 75 cm, the effective length is 65 cm) equipped with a special software for personal computers was applied. The amino acid composition of sapropel from Prybych deposits (Shatsky district, Volyn region) has been studied. The presence of a number of amino acids in sapropel with the total content of more than 2% has been found using capillary electrophoresis. Sapropel contains a vast amount of essential amino acids (histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan and valine) and a number of nonessential ones (alanine, arginine, glycine, proline and serine). Alanine, glycine, leucine, and isoleucine prevail in the mixture analyzed.*

It is known that the treatment by sapropel improves lymph and blood circulation, strengthens the vascular wall and stimulates the function of the autonomic nervous system. Sapropel stimulates metabolism in tissues, increases the oxygen exchange, has a tonic effect and pronounced antibacterial action, promotes activation of immune reactions, increases the body's resistance to diseases, successfully controls the pathogenic microflora of the mucous membranes, eliminates inflammation without damaging of the useful microflora, etc. [3, 4].

A diverse composition of sapropels allows to use them successfully in cosmetology. Sapropelic therapeutic muds exhibit anti-inflammatory and desensitizing effect, protect the body from the damaging effects of free radicals, slow down the aging processes, give the skin freshness and elasticity, improve the cell regeneration by 10%, moisturize and increase elasticity and thickness of the keratoid layer of the epidermis [4].

A wide range of indications for using sapropels demonstrates the importance of their application for prevention and treatment of diseases, and the relevance and prospects for their further studies to obtain new pharmaceuticals and cosmetics.

The aim of the study was to investigate the amino acid composition of sapropel of Prybych deposits (Shatsky district, Volyn region).

**Materials and Methods**

The study of anhydrous sapropel of Prybych deposits (Shatsky district, Volyn region) was carried out in the Laboratory of Feed Additives and Premixtures Control at the State Scientific-Research Control Institute of Veterinary Medicinal Products and Feed Additives in Lviv.

Currently for determination of amino acids chemical methods of research, the method of high performance liquid chromatography are used, as well as the method for separation of complex mixtures – capillary electrophoresis, which allows analyzing ionic and neutral components of various origin with a high efficiency; its application has increased within the last two decades [1, 11, 12].

Traditionally, capillary electrophoresis is compared with high performance liquid chromatography since in both methods separation takes place in a confined space (capillary or column) involving the mobile liquid phase (buffer or eluent), and for registration of signals the similar principles for detection and software data processing are applied [5, 9, 10].

The advantages of the capillary electrophoresis method are high efficiency of the sample separation; small amounts of the samples to be analyzed, buffers, highly pure and expensive organic solvents; the absence of columns and sorbents; short-term investigation periods [6, 7, 8].

The capillary electrophoresis method has been successfully applied for analysis of various substances (inorganic and organic cations and anions, amino acids, vitamins, dyes, proteins, etc.) and also for process control of production, incoming inspection of the raw material, analysis of pharmaceuticals and food products, in forensics, medicine, biochemistry, etc. [2].

The method is based on decomposition of the samples with acidic hydrolysis of amino acids into free forms of phenylisothiocarbamide derivatives (PTC-derivatives), their subsequent separation and quantitative determination by capillary electrophoresis.

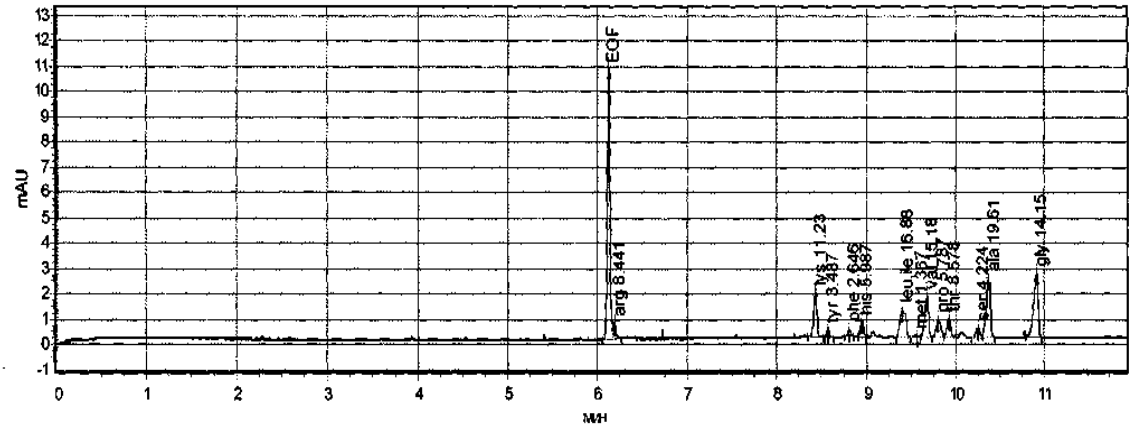


Fig. 1. The electrophoregram of acid hydrolysate of the test sample of sapropel No.1 (amino acids in the order of peaks: arginine, lysine, tyrosine, phenylalanine, histidine, leucine + isoleucine, methionine, valine, proline, threonine, serine, alanine, glycine).

In accordance with the guidelines of "Feed and feed raw material. Determination of amino acids by capillary electrophoresis using the capillary electrophoresis system "Kapel' – 105/105m" the method for determination of the mass fraction of such amino acids as arginine, lysine, tyrosine, phenylalanine, histidine, leucine and isoleucine (total), methionine, valine, proline, threonine, serine, alanine, and glycine was developed.

When conducting research by the capillary electrophoresis method the "Kapel' – 105/105 M" system with a positive polarity of a high voltage source (the internal diameter of the capillary is 50 μm, the full length of the capillary is 75 cm, the effective length is 65 cm) equipped with a special software for personal computers was applied.

When preparing for measurement the sampling, preparation of the capillary and calibrated solutions to work, calibration of "Kapel" system capillary electrophoresis and sample preparation were carried out.

The starting materials for preparing calibrated solutions are preparations of L-isomer of amino acids of guarantee purity (the content of the mass fraction of the active substance is at least 98%).

Performance of tests included the following successive stages: hydrolysis of samples, obtaining of PTC-derivatives of amino acids and analysis of the samples prepared.

*Hydrolysis of samples.* Place 100.0±0.2 mg of the weighed sample into a hydrolysis vial, add 10 cm³ of hydrochloric acid, seal and mix. Place the vial in the drying cabinet. Carry out hydrolysis at the temperature of 110°C for 14-16 hours. Remove vials with the samples from the drying cabinet after hydrolysis and cool to the room temperature. Cool and filter the content of vials through a "blue ribbon filter", reject the first portions and collect basic filtrates in a bowl with a lid to prevent evaporation. Then proceed to obtain PTC-derivatives.

*Obtaining of PTC-derivatives of amino acids.* Add 0.05 cm³ of the sample hydrolysates into 10-15 cm³ glass cups. Evaporate the solutions to dryness in the flow of warm air. Add 0.15 cm³ of sodium carbonate solution, 0.3 cm³ of phenyl isothioacetate (PTA) solution into each bottle with a dry residue. Thoroughly stir to dissolve the precipitate, close the lid and leave for 35 min at the room temperature. Then evaporate the solution to dryness in the flow of warm air.

*Analysis of the samples prepared.* Transfer solutions prepared for the analysis to Eppendorf test tubes, centrifuge for 1 minute at a speed of 6000 rev/min.

Record at least two electrophoregrams for each solution prepared in the appropriate conditions. Check the accuracy of the automatic marking of peaks after analysis. Using the software, conduct identification of the components in the sample comparing retention times of the components in the sample analyzed and the control mixture without exceeding the identification width more than 5%.

*Processing of the test results.* The mass fraction of each amino acid (X, %) was calculated by the formula:

$$X = \frac{10 \cdot C_{vis.}}{m},$$

where: $C_{vis}$ – is the value of the mass concentration of amino acid determined in solution, mg/dm³; *m* – is mass of the weighed sample, mg.

In parallel, two studies were performed. Record two *electrophoregrams* for each of the solutions prepared. The conditions of the study were the weighed sample of sapropel No.1 – 509.4 mg, sample No.2 – 300.1 mg, temperature – 30.0°C, the wavelength – 254 nm.

**Results and Discussion**

*Electrophoregrams* and the results of quantitative determination of amino acids are shown in Fig. 1, Fig. 2 and Tab. 1.

The results of the study are presented in Tab. 2 and show that sapropel contains essential amino acids (histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan and valine) and a number of nonessential amino acids (alanine, arginine, glycine, proline and serine), their total content is about 2%.

Alanine, glycine, leucine and isoleucine, and small amounts of serine, tyrosine, phenylalanine and methionine predominate in the mixture of amino acids studied.

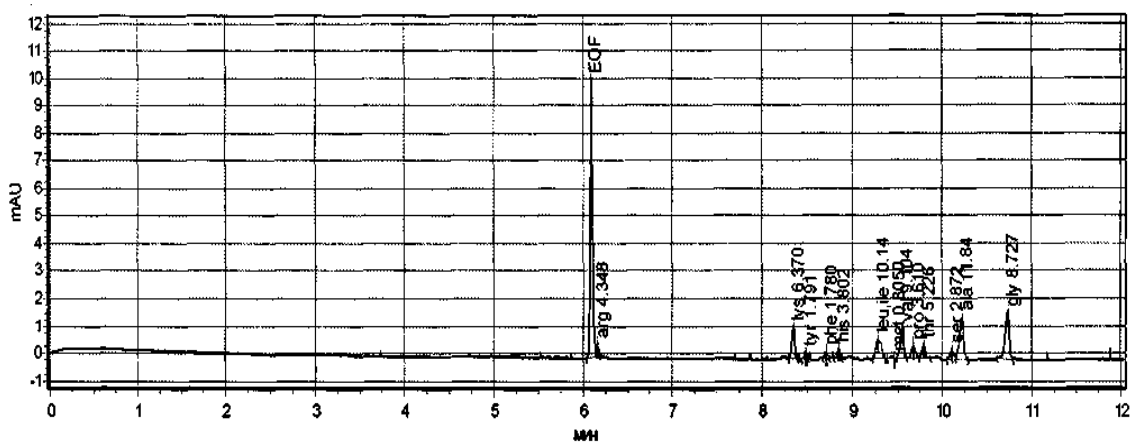


Fig. 2. The electrophoregram of acid hydrolysate of the test sample of sapropel No.2 (amino acids in the order of peaks: arginine, lysine, tyrosine, phenylalanine, histidine, leucine + isoleucine, methionine, valine, proline, threonine, serine, alanine, glycine).

Table 1

The results of quantitative determination of amino acids

| Amino acid | Retention time, sample 1, min | Peak area, sample 1, $cm^2$ | Concentration, sample 1, mg/l, $C_{vis}$ | Retention time, sample 2, min | Peak area, sample 2, $cm^2$ | Concentration, sample 2, mg/l, $C_{vis}$ |
|---|---|---|---|---|---|---|
| EOF | 6.123 | 200.4 | | 6.093 | 185.1 | |
| Arginine | 6.192 | 12.24 | 8.441 | 6.160 | 6.308 | 4.348 |
| Lysine | 8.433 | 44.40 | 11.23 | 8.352 | 25.18 | 6.370 |
| Tyrosine | 8.575 | 8.254 | 3.487 | 8.490 | 4.238 | 1.791 |
| Phenylalanine | 8.810 | 6.174 | 2.646 | 8.718 | 4.155 | 1.780 |
| Histidine | 8.948 | 13.65 | 5.987 | 8.852 | 8.670 | 3.802 |
| Leucine + isoleucine | 9.400 | 52.27 | 16.88 | 9.292 | 31.40 | 10.14 |
| Methionine | 9.558 | 3.654 | 1.367 | 9.463 | 2.151 | 0.8050 |
| Valine | 9.678 | 48.39 | 15.18 | 9.553 | 29.02 | 9.104 |
| Proline | 9.803 | 25.93 | 5.787 | 9.678 | 16.17 | 3.610 |
| Threonine | 9.923 | 22.72 | 8.578 | 9.793 | 13.84 | 5.226 |
| Serine | 10.248 | 13.71 | 4.224 | 10.107 | 9.324 | 2.872 |
| Alanine | 10.372 | 79.19 | 19.61 | 10.218 | 47.82 | 11.84 |
| Glycine | 10.908 | 87.12 | 14.15 | 10.732 | 53.73 | 8.727 |

Table 2

The quantitative analysis of amino acids in sapropel of Prybych deposits
$X \pm \Delta$, %, P = 0.95, n = 2

| Amino acid | Sample No.1 X,% | ±Δ, % | ±δ, % | Sample No.2 X,% | ±Δ, % | ±δ, % |
|---|---|---|---|---|---|---|
| Arginine | 0.14 | 0.06 | 40 | 0.16 | 0.06 | 40 |
| Lysine | 0.21 | 0.07 | 34 | 0.22 | 0.07 | 34 |
| Tyrosine | 0.06 | 0.02 | 30 | 0.07 | 0.02 | 30 |
| Phenylalanine | 0.06 | 0.02 | 30 | 0.05 | 0.02 | 30 |
| Histidine | 0.13 | 0.07 | 50 | 0.12 | 0.06 | 50 |
| leucine and isoleucine | 0.34 | 0.09 | 26 | 0.33 | 0.07 | 26 |
| Methionine | 0.03 | 0.01 | 34 | 0.03 | 0.01 | 34 |
| Valine | 0.3 | 0.12 | 40 | 0.3 | 0.12 | 40 |
| Proline | 0.12 | 0.03 | 26 | 0.11 | 0.03 | 26 |
| Threonine | 0.17 | 0.07 | 40 | 0.17 | 0.07 | 40 |
| Serine | 0.09 | 0.02 | 26 | 0.08 | 0.02 | 26 |
| Alanine | 0.39 | 0.10 | 26 | 0.38 | 0.09 | 26 |
| Glycine | 0.29 | 0.09 | 34 | 0.28 | 0.09 | 34 |

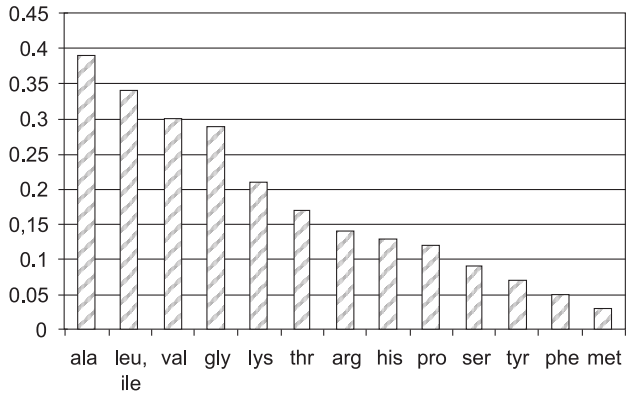


Fig. 3. The amino acids content in the sapropel samples analyzed.

The amino acids that are present in the mixture and their quantitative content (in %) are shown in Fig. 3.

CONCLUSIONS

1. The amino acid composition of sapropel from the Prybych lake deposits (Shatsky district, Volyn region) has been investigated. The presence of a number of essential and nonessential amino acids in sapropel with the total content of more than 2% has been found using capillary electrophoresis.

2. Sapropel contains a vast amount of essential amino acids (histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan and valine) and a number of nonessential ones (alanine, arginine, glycine, proline and serine). Alanine, glycine, leucine, and isoleucine prevail in the mixture.

REFERENCES

1. *Каменцев Я.С., Комарова Н.В. // Аналитика и контроль. – 2002. – Т. 6, №1. – С. 13-18.*
2. *Корми та кормова сировина. Визначення вмісту амінокислот методом капілярного електрофорезу з використанням системи капілярного електрофорезу «Капель-105/105М»: Метод. рекоменд. / Державний науково-дослідний контрольний інститут ветеринарних препаратів та кормових добавок. – Львів, 2013. – 47 с.*
3. *Куликов В.Ю. Адаптогенные и лечебные свойства пелоидов. – Новосибирск, 2001. – 219 с.*
4. *Струс О.Є. // Укр. журн. клін. та лабораторної медицини. – 2014. – Т. 9, №2. – С. 56-62.*
5. *Balammal G., Sekar B.M., Jayachandra R.P. // Intern. J. of Preclinical and Pharmac. Res. – 2012. – Vol. 3, Issue 1. – P. 50-63.*
6. *Brkljaca R., Urban S. // J. Liq. Chrom. Rel. Technol. – 2011. – Vol. 34. – P. 1063-1076.*
7. *Cordell G.A., Shin Y.G. // Pure Appl. Chem. – 1999. – Vol. 71. – P. 1089-1094.*
8. *Lambert M., Wolfender J.L., Staerk D. et al. // Anal. Chem. – 2007. – Vol. 379. – P. 727-735.*
9. *Lang G., Mayhudin N.A., Mitova M.I. et al. // J. Nat. Prod. – 2008. – Vol. 19. – P. 1595-1599.*
10. *Lewis R.J., Bernstein M.A., Duncan S.J., Sleigh C.J. // Magn. Reson. Chem. –2005. – Vol. 43. – P. 783-789.*
11. *Soga T. // Methods Mol. Biol. – 2007. – Vol. 358. – P. 129-137.*
12. *Sun Lin J., Mahyudin N.A., Chamyuang S. et al. // Proceedings of Manapro XII: 12th Intern. Symposium of Marine Natural Products, Feb 4th-9th, 2007, Queenstown, New Zealand. – Queenstown, 2007. – P. 130-138.*

**ДОСЛІДЖЕННЯ АМІНОКИСЛОТНОГО СКЛАДУ САПРОПЕЛЮ МЕТОДОМ КАПІЛЯРНОГО ЕЛЕКТРОФОРЕЗУ**

***О.Є.Струс***

***Ключові слова:*** *сапропель; метод капілярного електрофорезу; амінокислотний склад*

*Різноманітний склад сапропелів дозволяє успішно використовувати їх у косметології. Грязі лікувальні сапропелеві володіють протизапальною та десенсибілізуючою дією, захищають організм від руйнівної дії вільних радикалів, сповільнюючи процеси старіння, надають шкірі свіжості, пружності та еластичності, покращують клітинну регенерацію на 10%, зволожують та збільшують товщину і еластичність рогового шару епідермісу. Широкий спектр показань для використання сапропелів переконливо демонструє значимість їх використання у профілактиці та лікуванні захворювань, а також актуальність та перспективність їх подальшого дослідження з метою отримання нових лікарських та косметичних засобів. При проведенні досліджень методом капілярного електрофорезу використовували систему капілярного електрофорезу «Капель – 105/105 М» із позитивною полярністю джерела високої напруги (внутрішній діаметр капіляра 50 мкм, повна довжина капіляра – 75 см, ефективна довжина – 65 см), оснащену спеціальним програмним забезпеченням на основі персонального комп'ютера. Досліджено амінокислотний склад сапропелю родовища озера Прибич Шацького району Волинської області. Встановлено наявність у складі сапропелю низки амінокислот, сумарний вміст яких перевищує 2%. Сапропель містить переважну кількість незамінних амінокислот (гістидин, ізолейцин, лейцин, лізин, метіонін, фенілаланін, треонін, триптофан і валін ) і ряд замінних (аланін, аргінін, гліцин, пролін і серин) амінокислот. У суміші переважають аланін, гліцин, лейцин та ізолейцин.*

**ИССЛЕДОВАНИЕ АМИНОКИСЛОТНОГО СОСТАВА САПРОПЕЛЯ МЕТОДОМ КАПИЛЛЯРНОГО ЭЛЕКТРОФОРЕЗА**

***О.Е.Струс***

***Ключевые слова:*** *сапропель; метод капиллярного электрофореза; аминокислотный состав*

*Разнообразный состав сапропелей позволяет успешно использовать их в косметологии. Грязи лечебные сапропелевые обладают противовоспалительным и десенсибилизирующим действием, защищают организм от разрушительного воздействия свободных радикалов, замедляя процессы старения, придают коже свежесть, упругость и эластичность, улучшают клеточную регенерацию на 10%, увлажняют и увеличивают толщину и эластичность рогового слоя эпидермиса. Широкий спектр показаний для использования сапропелей убедительно демонстрирует значимость их использования в профилактике и лечении заболеваний, а также актуальность и перспективность их дальнейшего исследования с целью получения новых лекарственных средств. При проведении исследований методом капиллярного электрофореза использовали систему капиллярного электрофореза «Капель – 105/105 М» с положительной полярностью источника высокого напряжения (внутренний диаметр капилляра – 50 мкм, полная длина капилляра – 75 см, эффективная длина – 65 см), оснащенную специальным программным обеспечением на основе персонального компьютера. Исследован аминокислотный состав сапропеля месторождения озера Прибич Шацкого района Волынской области. Установлено наличие в составе сапропеля ряда аминокислот, суммарное содержание которых превышает 2%. Сапропель содержит подавляющее количество незаменимых аминокислот (гистидин, изолейцин, лейцин, лизин, метионин, фенилаланин, треонин, триптофан и валин) и ряд заменимых (аланин, аргинин, глицин, пролин и серин) аминокислот. В смеси преобладают аланин, глицин, лейцин и изолейцин.*

*Recommended by Doctor of Pharmacy, professor O.F.Piminov*

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# STABILITY ESTIMATION OF THE WOOL FAT SUBSTANCE WHEN STORING IN THE PHARMACY

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*Key words: stability; ointment bases; wool fat; extemporal compounding*

*One of the main objectives for drug manufacturers is to study stability, which is the quality factor for any drug. The requirements of stability are imposed to the extemporaneous medicines too. The Guide for the Compounding Practitioner (USA) contains article "1191" "Stability considerations in dispensing practice" with recommendations to repacking of substances, which can be used in pharmaceutical compounding. The pharmacy receives the wool fat substance in the original packing with a large volume that causes the necessity of its opening and the substance repacking to the container of a smaller volume for direct use. The analysis of the wool fat substance stability under conditions of its repacking from original packing to the amber glass container during the research has been done. The research has shown its compliance with the parameters set. The repacked substance was stored at the temperature of + 25°C in the pharmacy conditions in the assistant room within four months. Re-analysis of the quality parameters indicates the compliance with the requirements of the "Quality control methods" (QCM) and the European Pharmacopoeia. It has been found that the acid and peroxide values have increased only a little, and the water absorption ability of the substance has decreased. The results obtained have shown that all quality parameters of the wool fat substance in relation to stability when storing in the pharmacy after its repacking from original packing to the amber glass container are preserved within four months.*

Stability is one of the main parameters of the drug quality, it provides preservation of its therapeutic and preventive properties during its storage period [2, 6, 7]. One of the main stages of drug manufacturing is the study of stability of drugs and determination of their expiration date [6].

Stability is the factor of the drug quality. The literal meaning of stability is the ability of a drug product to remain within the quality specifications to provide its identity, strength, and purity. Stability comes into focus when the quality, efficiency and safety of the drug are concerned. The main objective of the stability study is to determine its sensitivity to the different environmental factors and assess the shelf-life of the drug and the storage conditions recommended [4, 8, 9].

The requirements of stability to the extemporaneous medicines are contained in article "1191" "Stability considerations in dispensing practice" of the Guide for the Compounding Practitioner [10] developed on the basis of the US Pharmacopoeia requirements. It has the subsection "Responsibility of Pharmacists", which describes indicators of instability of all extemporaneous dosage forms. For semisolids the primary indication of instability is often either discoloration or a noticeable change in consistency or odour. The common signs of instability of ointments are the change in consistency and separation of excessive amounts of liquid and formation of granules or granularity [3, 10].

Stability of the base considerably influences on the quality of the dosage form. In the prescriptions of the compounding ointments wool fat is used as a base (with White soft paraffin or as an independent base). The pharmacy receives the substance in the original packing and after its opening wool fat is stored in the amber glass or ceramic containers.

The aim of this work is to assess stability of the wool fat substance for compounding ointments within four months when storing in the amber glass containers in the pharmacy conditions.

**Materials and Methods**

For our research the wool fat substance manufactured by "Lanolines Stella S. A.", Belgium; the amber glass containers; tableware and reagents corresponding to the requirements of the State Pharmacopoeia of Ukraine were used. The experimental work was carried out in the laboratory of the quality control of medicines of the State inspectorate of the quality control of medicines in the Donetsk region.

*Quality parameters of wool fat*

*Drop point* [1, 5]: 38ºC to 44ºC. Place the test sample into a metal cup, melt the wool fat on a water-bath, cool to about 50ºC, pour into the cup and allow to stand at 15-20ºC for 24 h.

*Water absorption ability* – Place 10 g of the molten wool fat into a mortar and allow to cool to the room temperature. Weight the mortar. Add water *R* in portions of 0.2-0.5 ml from a burette stirring vigorously after each addition to incorporate water *R*. Instead of a pestle, use a high-density polypropylene cylindrical rod. The end-point is reached when visible droplets remain and cannot be incorporated. Weight the mortar again and determine the amount of water absorbed by the weight difference. Not less than 20 g of water *R* should be absorbed [5].

Table

The results of stability studies of the wool fat substance during its storage period

| Quality parameter | The research conducted* | | Requirements | Results |
|---|---|---|---|---|
| | Experiment 1 | Experiment 2 | | |
| Description | + | + | A yellow pasty substance, when melted it is a clear or almost clear, yellow liquid. The solution in light petroleum is opalescent (QCM, EP) | satisfied |
| Drop point | 44°C | 44°C | 38-44°C (QCM, EP) | satisfied |
| Water absorption ability | 29.2 ml | 23.0 ml | not less than 20 ml (QCM, EP) | satisfied |
| Acid value | 0.57 | 0.60 | maximum 1.0 (QCM, EP) | satisfied |
| Peroxide value | 12.0 | 16.6 | maximum 20 (QCM, EP) | satisfied |
| Saponification value | 90.7 | 90.8 | 90 to 105 (QCM, EP) | satisfied |
| Loss on drying | 0.25% | 0.27% | maximum 0.5% (QCM, EP) | satisfied |
| Sulphated ash | 0.03% | 0.03% | maximum 0.15% (QCM, EP) | satisfied |
| Paraffines | absent | absent | not more than 1.0% (QCM, EP) | satisfied |
| Chlorides | absent | absent | not more than 150 ppm (QCM, EP) | satisfied |

* – the average result of three measurements is given for each experiment

*Acid value* – not more than 1.0. Dissolve 5.0 g of the test sample in 25 ml of the mixture of equal volumes of ethanol *R* and light petroleum *R3* previously neutralized with 0.1 M potassium hydroxide [1, 5].

*Peroxide value* [1, 5] – not more than 20. Before adding 0.5 ml of saturated potassium iodide solution *R* cool the solution obtained to the room temperature.

*Saponification value* [1, 5] – from 90 to 105. Heat 2.00 g of the test sample under reflux for 4 h.

*Paraffines* [5] – not more than 1.0%. The stopper and cotton plugs used must be free from lubricants. Prepare a column with anhydrous aluminium oxide of 0.23 m long and 20 mm in diameter by adding a suspension of anhydrous aluminium oxide *R* and light petroleum *R1* to a glass tube fitted with a stopper (before use, dehydrate the anhydrous aluminium oxide by heating it in an oven at 600 ºC for 3 h). Allow to settle and reduce the depth of the layer of the solvent above the column to about 40 mm. Dissolve 3.0 g of the substance to be examined in 50 ml of warm light petroleum *R1*, cool, pass the solution through the column at the flow rate of 3 ml/min and wash with 250 ml of light petroleum *R1*. Concentrate the combined eluate and the washing liquid to the small volume by distillation, evaporate to dryness on a water bath and heat the residue at 105ºC for 10 min until the difference between two successive weighings will not exceed 1 mg. The weight of the residue should not exceed 30 mg.

*Chlorides* [5] – the content of chlorides does not exceed 150 ppm. Boil 1.0 g of the substance with 20 ml of ethanol *R* (90 % v/v) in a round-bottomed flask fitted with a reflux condenser for 5 min. Cool, add 40 ml of water *R* and 0.5 ml of nitric acid *R* and filter. To the filtrate add 0.15 ml of 10 g/l solution of silver nitrate *R* in ethanol *R* (90 % v/v). Allow to stand for 5 min protected from light. Opalescence in the test solution should not be more intense than that in the standard solution prepared at the same time by adding 0.15 ml of 10 g/l solution of silver nitrate *R* in ethanol *R* (90 % v/v) to the mixture of 0.2 ml of 0.02 M hydrochloric acid, 20 ml of ethanol *R* (90 % v/v), 40 ml of water *R* and 0.5 ml of nitric acid *R*.

*Loss on drying* [5] – not more than 0.5 %. Dry 1.000 g of the substance under study in a drying cabinet at 105ºC for 1 h.

*Sulphated ash* [5] – not more than 0.15 %. Ignite 5.0 g of the test sample and use the residue to determine the sulphated ash.

**Results and Discussion**

The subsection "Responsibility of Pharmacists" of the article "1191" "Stability considerations in dispensing practice" of the Guide for the Compounding Practitioner (USA) [3, 10] contains recommendations about repacking of substances, which can be used in pharmaceutical compounding. In general, repacking is inadvisable. If repacking is necessary, it is essential to use suitable containers. First of all, it must be made of a neutral material.

If stability data of the substance after its repacking to the new container are not available, repacking only the necessary quantities for use in a short period of time is recommended. On the label of the new package a series of substance and the expiration date must be specified. If a sterile product is repacked from a multiple-dose vial into unit-dose syringes, discard the latter if it is not used within 24 hours unless the data are available to support longer storage. If quantities are repacked in advance of immediate need, maintain suitable repacking records indicating the name of the manufacturer, the lot number, date and persons that are responsible for repacking and checking. If safety closures are required, use container closure systems that ensure compliance with compendial and regulatory standards for storage.

The pharmacies receive the wool fat substance in the original packing with a large volume that causes the necessity of its opening and the substance repacking to the container of a smaller volume for direct use and subsequent storage. Since in the literature there is no

information about stability of the wool fat substance after its repacking from original packing, the aim of our further work was to study the stability parameters of the given substance.

Part of the wool fat substance from the original packing was bought for the research and dispensed into the amber glass containers. The analysis of the substance carried out in a week after repacking has shown its compliance with the QCM requirements (Table, experiment 1) containing the list of tests given to verify the quality of the wool fat substance in the European Pharmacopoeia [5].

After that wool fat was stored at temperature of + 25°C in the assistant room. The second analysis of the wool fat substance compliance with all quality parameters according to the QCM was conducted in four months (Table, experiment 2). The results obtained indicate the preservation of stability of the wool fat substance when storing in the pharmacy conditions in the amber glass container. Only the acid value (from 0.57 to 0.60) and the peroxide value (from 12.0 to 16.6) slightly increased a little, and the water absorption ability of the wool fat substance (from 29.2 ml to 23 ml) decreased. However, all quality parameters of the wool fat substance correspond to the parameters set in QCM and do not exceed them.

CONCLUSIONS

1. Stability of the wool fat substance during its storage in the amber glass container in the pharmacy conditions within four month has been studied.

2. During the period studied the water absorption ability of the wool fat decreased, and the acid and peroxide values e increased. However, all parameters are in compliance with the QCM requirements.

3. The results obtained have shown the possibility of repacking and storage of the wool fat substance in the amber glass containers in the pharmacy conditions.

4. Our further research will be devoted to the study of the microbiological purity of the bases to verify their compliance with the requirements set.

REFERENCES


1. *Державна фармакопея України / Державне підриємствово «Науково-експертний фармакопейний центр». – 1-е вид. – Х.: РІРЕГ, 2001. – 556 с.*
2. *Мешковский А.П. // Аптека. – 2000. – №236 (15) [Electronic resource]. – Access mode: http://www.apteka.ua/article/10636.*
3. *Allen V.L. // Secundum artem. – Vol. 7, №3 [Electronic resource]. – Access mode : http://drofrx.com/Compounding_files/Secundum%20Artem/Sec%20Artem%207.3.pdf.*
4. *Bajaj S., Singla D., Sakhuja N. // J. of Applied Pharm. Sci. – 2012. – Vol. 02 (03). – P. 129-138.*
5. *European Pharmacopoeia. – 6-th ed. – Strasbourg: European Directorate for the Quality of Medicines and HealthCare of the Counsil of Europe, 2008. – Vol. 2.2. – P. 3222-3226.*
6. *Huynh-Ba K. Handbook of stability testing in pharmaceutical development. – New York: Springer, 2009. – 406 p.*
7. *Kupiec T.C., Skinner R., Lanier L. // Int. J. of Pharm. Comp. – 2008. – Vol. 12, №1. – P. 50-53.*
8. *Panda A., Sukhadakulkarni, Tiwari R. // Int. J. of Pharm. Res. and Biosci. – 2013. – Vol. 2 (6). – P. 69-80.*
9. *Quality assurance of pharmaceuticals: a compendium of guidelines and related materials. – Geneva: World Health Organization, 2007. – 2-nd ed. – Vol. 2. – 409 p.*
10. *USP compounding: A Guide for the Compounding Practitioner / The United State Pharmacopoeia convention [Electronic resource]. – Rockville: The United States Pharmacopeial Convention, 2012. – 317 p.*


**ОЦІНКА СТАБІЛЬНОСТІ СУБСТАНЦІЇ ЛАНОЛІНУ ПРИ ЗБЕРІГАННІ В УМОВАХ АПТЕКИ**

***Л.П.Савченко, М.О.Хмельова, О.А.Євтіфєєва, В.А.Георгіянц***

***Ключові слова:*** *стабільність; мазеві основи; ланолін безводний; аптечне виготовлення*

*Однією з основних задач, яка стоїть перед виробником лікарського засобу, є дослідження його стабільності. Вона є одним із факторів якості препарату. Вимоги до збереження стабільності висуваються і до екстемпоральних лікарських засобів. До Керівництва для аптечних працівників США входить стаття «1191» «Поняття стабільності в аптечній практиці», в якій містяться рекомендації щодо перепакування субстанцій, які можуть використовуватись в аптечній практиці. Субстанція ланоліну надходить до аптечного закладу в заводських упаковках. Оскільки їх об'єм досить великий, необхідним є розфасування субстанції в тару меншого об'єму для безпосереднього використання. В процесі дослідження здійснено оцінку стабільності субстанції ланоліну безводного при його перепакуванні з заводської тари в аптечну скляну тару з темного скла. Проведений аналіз за всіма показниками якості субстанції ланоліну безводного свідчить про її відповідність встановленим параметрам. Розфасована субстанція зберігалась при температурі + 25°С в умовах аптеки в асистентській кімнаті впродовж чотирьох місяців. Повторне дослідження показників її якості свідчить про відповідність «Методам контролю якості» (МКЯ) та вимогам Європейської Фармакопеї (ЄФ). Встановлено, що дещо збільшились лише кислотне та пероксидне число, а також зменшилась водоабсорбційна здатність субстанції. Отримані результати свідчать про збе-*

*реження показників якості щодо стабільності субстанції ланоліну при його перефасуванні та зберіганні в умовах аптеки в штангласах з темного скла впродовж чотирьох місяців.*

**ОЦЕНКА СТАБИЛЬНОСТИ СУБСТАНЦИИ ЛАНОЛИНА ПРИ ХРАНЕНИИ В УСЛОВИЯХ АПТЕКИ**

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***Ключевые слова:*** *стабильность; мазевые основы; ланолин безводный; аптечное приготовление*

*Одной из основных задач, которая стоит перед производителем лекарственного средства является исследование его стабильности. Она является одним из факторов качества препарата. Требования к сохранению стабильности выдвигаются и к экстемпоральным лекарственным средствам. В Руководство для аптечных сотрудников США входит статья «1191» «Понятие стабильности в аптечной практике», в которой содержатся рекомендации о переупаковке субстанций, которые могут использоваться в аптечной практике. Субстанция ланолина поступает в аптечное учреждение в заводских упаковках. Поскольку их объем достаточно большой, необходимым является ее расфасовка в тару меньшего объема для непосредственного использования. В процессе исследования осуществлена оценка стабильности субстанции ланолина безводного после его переупаковки с заводской тары в аптечную стеклянную тару из темного стекла. Проведенный анализ по показателям качества субстанции ланолина безводного свидетельствует о ее соответствии установленным параметрам. Расфасованная субстанция хранилась при температуре + 25°С в условиях аптеки в ассистентской комнате на протяжении четырех месяцев. Повторное исследование показателей ее качества свидетельствует о соответствии «Методам контроля качества» (МКК) и требованиям Европейской Фармакопеи (ЕФ). Установлено, что несколько увеличились показатели только кислотного и пероксидного чисел, а также уменьшилась водоабсорбционная способность субстанции. Полученные результаты свидетельствуют о сохранении показателей качества в отношении стабильности субстанции ланолина при его перефасовке и хранении в условиях аптеки в штанглассах из темного стекла на протяжении четырех месяцев.*

# ТЕХНОЛОГІЯ ЛІКАРСЬКИХ ПРЕПАРАТІВ

*Recommended by Doctor of Pharmacy, professor N.P.Polovko*

UDC 615.014.22:615.454.1:616.211-002.193

## DEVELOPMENT OF THE TECHNOLOGY OF "FITORYN-PLUS" NASAL GEL FOR TREATING ALLERGIC RHINITIS

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*Key words: technology; nasal gel; allergic rhinitis*

*There are some available synthetic drugs of the foreign origin among symptomatic nasal medicines at the current pharmaceutical market. Most of them are produced in the form of drops or sprays, and it causes short duration of action of these drugs. Nowadays nasal gels attract great attention to the treatment of allergic rhinitis. The aim of the work is to develop a new nasal medicine based on biologically active plant substances in the gel form under the conditional name "Fitoryn-plus". A dry extract of licorice root and essential oils of eucalyptus and Siberian pine were used as active ingredients of this nasal gel. In order to choose a structure-forming component of the gel base 9 model samples of gel systems were prepared, and the possibility of using carbomer (carbopol) 934, hydroxyethyl cellulose and sodium alginate was investigated. To neutralize aqueous dispersions of carbopol the ammonia solution, the solution of sodium hydroxide and triethanolamine were used. The gel was prepared at the room temperature using two variants of technology: by mixing the finished gel and solutions of active substances previously prepared (technology 1) and by introducing active substances into the dispersion medium before the gel thickening (technology 2). Based on the complex of organoleptic, physical and chemical, structural and mechanical studies conducted a gelling agent, a neutralizing agent, a hydrophilic nonaqueous solvent and their optimal concentrations in the drug composition have been chosen. It has been shown that thermal effects of the samples of the active substances, the gel base and "Fitoryn-plus" gel are similar in nature; it may be subjectively indicative of the absence of chemical interaction between the components of the drug and validate the technology developed.*

Allergic rhinitis is a disease of the nasal epithelial membranes and is characterized by episodic disturbance of nasal breathing. Despite the fact that rhinitis is perceived by many patients as a temporary phenomenon, in doctors' opinion, it is the initial manifestation of systemic allergy.

Moreover, if this pathology is not treated, a chain of pathological processes in the lower respiratory system will start; and it can lead to development of some severe diseases such as asthma. Therefore, it is necessary to treat allergic (intermittent) rhinitis since the appearance of the first symptoms of this pathology [8, 10].

According to the World Health Organization, over the last ten years the number of patients suffering from allergic rhinitis has increased approximately twice, indicating the epidemic nature of the disease [5, 9].

The first symptoms that characterize intermittent rhinitis are formed from a few seconds to twenty minutes after direct interaction with the allergen. Patients suffering from allergic nasal pathology complain of nasal congestion and severe watery discharge from the nasal passages.

The main signs of allergic rhinitis include the following symptoms: frequent sneezing, itchy nose, scratchy throat, swelling of the face, watery eyes, headaches, hearing and smell disorders, etc. At the first signs of the disease the systemic and symptomatic treatment is needed for adults and children [3, 7].

There are some available synthetic drugs of the foreign origin among symptomatic nasal medicines at the current pharmaceutical market. Most of them are produced in the form of drops or sprays, and it causes short duration of action of these drugs [2].

Nowadays nasal gels attract great attention to the treatment of allergic rhinitis; they provide retention of the active substances on the nasal mucosa, do not disrupt the movement of ciliated epithelium, maintain the natural moisture of the nasal mucosa and provide a prolonged therapeutic action [4, 6].

Therefore, the aim of our work is to develop a new nasal medicine based on biologically active plant substances in the gel form under the conditional name "Fitoryn-plus".

**Materials and Methods**

A dry extract of licorice root and essential oils of eucalyptus and Siberian pine were used as active ingredients of the nasal gel proposed. Taking into account their solubility in well known solvents this dry extract of licorice root was introduced into the gel composition as an aqueous solution, and essential oils were as a solution in ethanol (96%).

Table 1

The composition of model gel bases (%)

| Model gel base | Carbopol 934 P | Hydroxyethyl-cellulose | Sodium alginate | Triethanolamine | Propylene glycol | Purified water |
|---|---|---|---|---|---|---|
| 1 | 0.5 | – | – | 0.5 | 10.0 | up to 100.0 |
| 2 | 1.0 | – | – | 1.0 | 10.0 | up to 100.0 |
| 3 | 1.5 | – | – | 1.5 | 10.0 | up to 100.0 |
| 4 | – | 1.0 | – | – | 10.0 | up to 100.0 |
| 5 | – | 1.5 | – | – | 10.0 | up to 100.0 |
| 6 | – | 2.0 | – | – | 10.0 | up to 100.0 |
| 7 | – | – | 5.0 | – | 10.0 | up to 100.0 |
| 8 | – | – | 7.0 | – | 10.0 | up to 100.0 |
| 9 | – | – | 10.0 | – | 10.0 | up to 100.0 |

In order to choose a structure-forming component of the gel base 9 model samples of gel systems were prepared, and the possibility of using carbomer (carbopol) 934, hydroxyethyl cellulose and sodium alginate as a gelling agent was investigated.

The solutions of ammonia, sodium hydroxide, as well as triethanolamine were used to neutralize the aqueous dispersions of carbopol. The appearance and colour of the model gel samples were estimated in the course of the experiment.

The gel was prepared at the room temperature using two variants of technology: by mixing the finished gel and the solutions of active substances previously prepared (technology 1) and by introducing active substances into the dispersion medium before the gel thickening (technology 2).

Determination of homogeneity of the samples was performed by the method of the State Pharmacopoeia of Ukraine (SPhU) [1]. The appearance, colour and odour was determined in accordance with the State Standard – GOST 29188.90.

The colloidal structure and thermal stability of the gel were visually assessed by the method of GOST 29188.3-91 at the room temperature (15-25°C), at 40°C and 5°C, as well as after cycles of freezing and thawing.

Determination of pH of aqueous extracts of the drug was conducted by the potentiometric method according to the SPhU [1].

Rheological studies were performed on a BROOKFIELD DV-II + PRO viscometer (USA) with the system of coaxial cylinders. Structural and mechanical properties were determined compared to "Loryzan" nasal gel widely used in the treatment of allergic rhinitis.

Water absorption kinetics of the drug was determined in experiments *in vitro* using the method of dialysis through a semipermeable membrane at (37.0±0.1)°C by changing the mass of the chamber with the sample.

Thermogravimetric analysis was performed on a Q-1000 derivatograph of the system of F.Paulik, J.Paulik, L.Efdey using the method of the SPhU [1]. Curves T (temperature), TG (change in weight), DTA (differential curve of thermal effects change) and DTG (differential curve of weight change) were recorded. As a reference aluminum oxide powder was used. The weight of the sample was 200 mg.

**Results and Discussion**

The optimal structure-forming component was selected on the basis of studies of physical and chemical stability of the gel model samples (Tab. 1). As a result of the studies conducted it has been found that model systems 1, 6 and 9 have too thin or, on the contrary, dense consistency, poorly applied to the mucosa and are almost not absorbed.

Model gel bases 2, 3, 7 and 8 both after preparation and after keeping for 30 days at 5°C, 20°C, 40°C and after 5 cycles of freezing / thawing (the temperature range was from -10°C to +45°C) were stable.

Samples of bases 4 and 5 did not withstand tests on colloidal and thermal stability. Therefore, for further study the gel systems 2, 3, 7 and 8 were chosen.

These gel samples were packed for storage at two temperature conditions – 8-15°C and 15-25°C. The results of the stability study of the bases developed while storing (within 30 days) are given in Tab. 2.

As it is seen from the data of Tab. 2, model samples 7 and 8 while storing for 30 days appeared to be un-

Table 2

The results of the stability study of gel samples 1, 2, 7 and 8 while storing

| No. | Colour | Odour | Homogeneity | Colloidal stability (visually) | Thermostability (visually) |
|---|---|---|---|---|---|
| 2 | brown | pleasant | homogeneous | stable | stable |
| 3 | brown | pleasant | homogeneous | stable | stable |
| 7 | appearance of a black stain | specific | homogeneous | unstable | unstable |
| 8 | appearance of a black stain | specific | homogeneous | unstable | unstable |

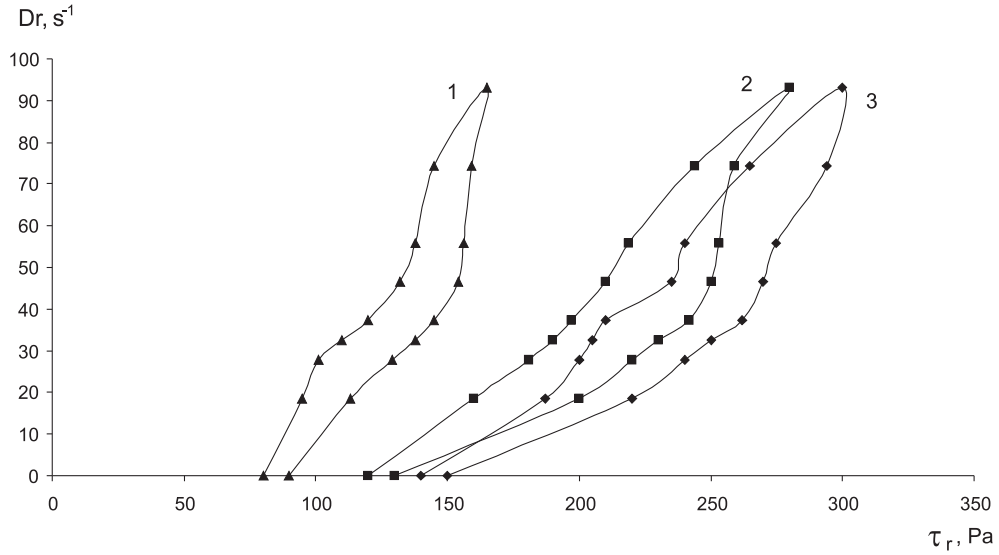


Fig. 1. Flow rheograms of the gel studied: sample 2-1, sample 3-2, and "Loryzan" gel – 3 at the temperature of (20.0±1.0)°C.

stable. On the 15-th day of storage the appearance of a black stain was observed, indicating the microbial contamination of the bases developed. In addition, these samples of gel bases did not withstand tests on colloidal and thermal stability while storing. For further research the model gel samples 2 and 3 were selected.

When carrying out rheological studies of these samples it has been found that while increasing of the carbopol concentration in the systems studied there is a shift from plastic to pseudoplastic type of flow and the appearance of thixotropic properties.

Model samples of gel systems containing the gelling agent in the concentration of more than 1.5% had a dense heterogeneous gel-like structure; their further neutralization led to a sharp increase in the structural viscosity, formation of a dense gel, in which the active ingredients could not be introduced and distributed evenly.

Neutralization of aqueous dispersions of carbopol caused increase of rheological parameters and formation of transparent gels. The effectiveness of gelation (increase of the structural viscosity after neutralization) depended on the concentration of carbopol.

In the range of 1.0-1.5% gelation was more effective. With increase of the carbopol concentration in more than 1.5 % the structural viscosity increased slightly, confirming the feasibility of preparing a gel within the concentration range of 1.0-1.5% (Fig. 1).

As it is shown in Fig. 1, sample 3 has rheological parameters most closely resembling rheological parameters of "Loryzan" nasal gel, and it has been also confirmed by the calculated values of mechanical stability and dynamic dilution ratios (1.08 and 1.05, 72% and 70.5%, respectively).

Sample 2 is more liquid, and its width of "hysteresis loop" indicates the less distinct thixotropic properties.

In further research when choosing a neutralizing agent to the gel system 3 the solutions of ammonia and sodium hydroxide, as well as triethanolamine were introduced. Samples neutralized with the ammonia solution and the sodium hydroxide solution darkened over time. While using the ammonia solution salting-out was also observed. Therefore, triethanolamine was used further since its addition did not change physical and chemical properties of the gel during the experiment. The optimal concentration of the neutralizing agent in the drug composition was determined when studying pH of its model samples (Tab. 3).

Table 3

The results of studying pH of the model samples of "Fitoryn-plus" gel

| No. | The content of triethanolamine | pH value |
|---|---|---|
| 3-a | 1.0% | 5.85±0.05 |
| 3-b | 1.5% | 6.40±0.05 |

As it can be seen from the table data, the model sample of the drug containing 1.5% of triethanolamine has the pH value that is the most close to the normal pH value of the human nasal mucosa, which is 6.0-7.0.

Propylene glycol was introduced to the composition of the nasal drug studied in order to provide a moderate osmotic activity, prevent dryness and irritation of the nasal mucosa. In addition, it is known that a positive moment of introduction of hydrophilic nonaqueous solvents into gels provides stability of their composition during the technological process.

We prepared model samples of gels containing propylene glycol from 10% to 30% and studied the kinetics of water absorption of these samples in quintuplicate. The concentration 10% was identified as the optimal concentration of propylene glycol (Fig. 2).

As it is shown in Fig. 2, the sample of "Fitoryn-plus" gel containing 10% of propylene glycol provides a moderate osmotic activity of the drug for 7 h, and it may indicate the absence of the irritating action. With introduction of propylene glycol in higher concentrations the osmotic activity of the samples reached 60-75%. It is not acceptable for nasal gels from the biomedical point of view.

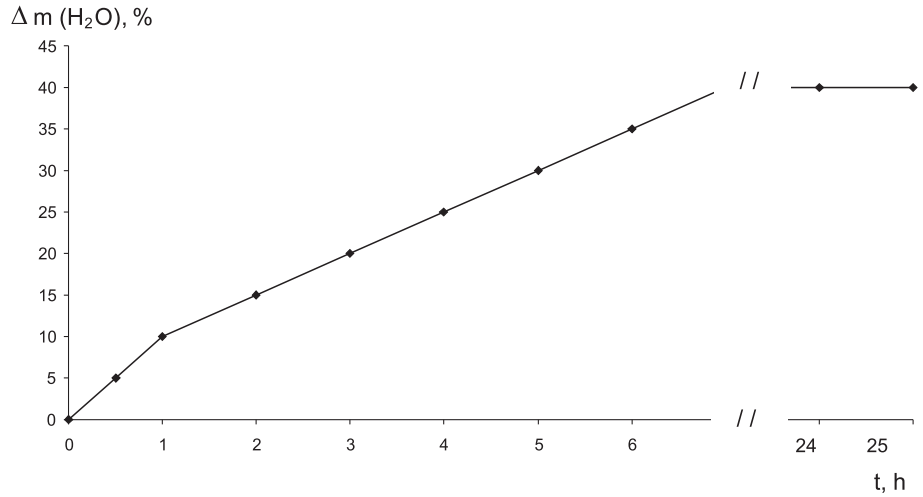


Fig. 2. The osmotic activity of "Fitoryn-plus" nasal gel containing 10% of propylene glycol.

Table 4

Physical and chemical properties of the samples of "Fitoryn-plus" gel

| Medicine | Colour | Odour | Homogeneity | pH | Colloidal stability | Thermal stability |
|---|---|---|---|---|---|---|
| Gel by technology 1 | brown | specific pleasant | + | 6.70±0.05 | + | + |
| Gel by technology 2 | brown | specific pleasant | + | 6.65±0.05 | - | - |

Considering all experimental data the samples of "Fitoryn-plus" gel by technology 1 and 2 were prepared.

The gels obtained by the technologies proposed had gel-like homogeneous consistency with a specific pleasant odour, brown colour, pH = 6.0-7.0. The study of thermal and colloidal stability proved stability of the system prepared only by technology 1 (Tab. 4).

Therefore, technology 1 (introduction of active ingredients to the finished gel) was determined as the rational technology for preparing "Fitoryn-plus" gel.

Then the thermogravimetric studies were conducted with the samples of the active ingredients, the gel base and the gel developed. When analyzing thermogravimetric curves of these samples it has been found that essential oils of Siberian pine and eucalyptus are stable up to the temperature of (50.0±1.0)°C, and at the temperatures ranging from 53°C to 84°C their losses in mass are up to 3%, the process of the sample destruction ends at the temperature of 200°C.

A dry extract of licorice root is stable up to the temperature of (37.0±1.0)°C, within the temperature range from 37°C to 130°C there is a gradual loss in its mass (Fig. 3).

The base starts melting at the temperature of (37.0± ±1.0)°C.

The process of decomposition of the gel takes place in two stages (Fig. 4). At the first stage the significant moisture loss in weight is not observed up to 37°C. The second (37-100)°C stage is characterized by the rapid

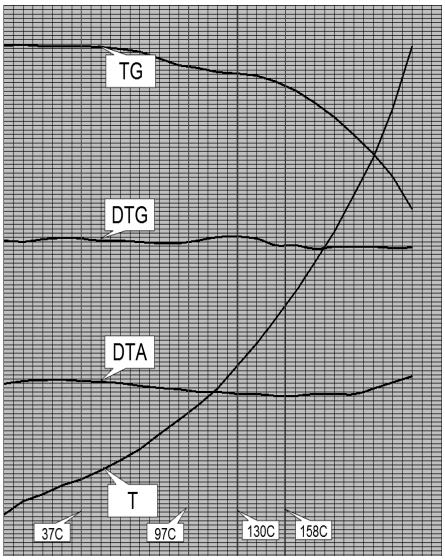


Fig. 3. Thermogravimetric curves of a dry extract of licorice root.

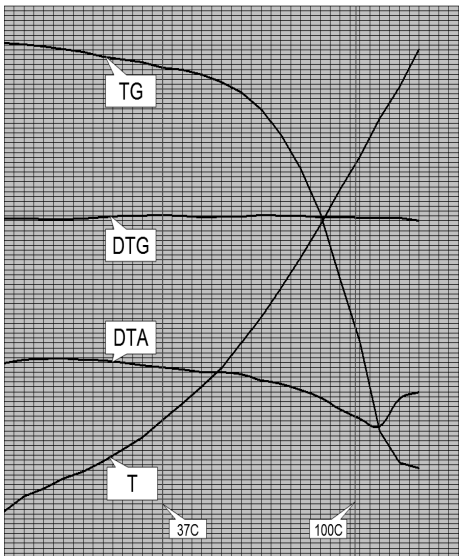


Fig. 4. Thermogravimetric curves of "Fitoryn-plus" gel.

and continuous process of destruction accompanied by significant exothermic effects.

Therefore, based on the thermogravimetric studies conducted it has been found that thermal effects of the samples are similar in nature; it may indicate a subjective absence of chemical interaction between components of the gel prepared according to the technology proposed.

CONCLUSIONS

1. The rational technology of the nasal gel under the conditional name "Fitoryn-plus" for treating allergic rhinitis has been developed.

2. The complex of organoleptic, physical and chemical, structural and mechanical studies was conducted; based on it a gelling agent, a neutralizing agent, a hydrophilic nonaqueous solvent and their optimal concentrations in the drug composition have been chosen.

3. It has been shown that thermal effects of the samples of the active substances, the gel base and "Fitoryn-plus" gel are similar in nature; it may be subjectively indicative of the absence of chemical interaction between the components of the drug and validate the technology developed.

REFERENCES


1. *Державна фармакопея України / Державне підприємство «Науково-експертний фармакопейний центр». – 1-е вид., 2 доп. – Х.: РІРЕГ, 2008. – 620 с.*
2. *Компендиум 2012 – лекарственные препараты / Под ред. В.Н.Коваленко. – К.: Морион, 2012. – http:// compendium.com.ua.*
3. *Косаковский А.Л., Гавриленко Ю.В. // Укр. мед. часопис. – 2013. – №6(98). – С. 23-27.*
4. *Angiera E., Willingtonb J., Scaddingc G. et al. // Primary Care Respiratory J. – 2010. – Vol. 19, №3. – P. 217-222.*
5. *Chowdhury M.A., Golam Rabbani S.M., Yasmeen N., Malakar M. // Bangladesh J. of Otorhinolaryngol. – 2010. – Vol. 16, №1. – P. 44-47.*
6. *Demoly P., Calderon M., Casale T. et al. // Clinical and Translational Allergy. – 2013. – Vol. 3, №7. – P. 84-92.*
7. *Kennedy J. // Self Care. – 2011. – Vol. 2, №2. – P. 55-59.*
8. *Shah A., Pawankar R. // Asian Pacific J. of Allergy and Immunol. – 2009. – Vol. 27, №1. – P. 71-77.*
9. *Small P., Kim H. // Allergy, Asthma & Clinical Immunol. – 2011. – Vol. 7, №1. – P. 125-133.*
10. *Williams D.C., Edney G., Maiden B. // World Allergy Organization J. – 2013. – Vol. 6, №4. – P. 331-337.*


**РОЗРОБКА ТЕХНОЛОГІЇ НАЗАЛЬНОГО ГЕЛЮ «ФІТОРИН-ПЛЮС» ДЛЯ ЛІКУВАННЯ АЛЕРГІЧНОГО РИНІТУ**

***О.А.Рухмакова, Т.Г.Ярних***

***Ключові слова:*** *технологія; назальний гель; алергічний риніт*

*На сучасному фармацевтичному ринку серед симптоматичних назальних засобів наявними є переважно синтетичні лікарські препарати закордонного виробництва. Значна їх частина випускається у формі крапель або спреїв, що обумовлює короткочасність дії. Все більшу увагу на сьогоднішній день у лікуванні алергічного риніту привертають назальні гелі. Метою роботи є розробка назального засобу на основі речовин рослинного походження у формі гелю під умовною назвою «Фіторин-плюс». В якості діючих речовин було використано сухий екстракт кореня солодки та ефірні олії сосни сибірської та евкаліпту. З метою вибору структуроутворюючого компонента досліджена можливість використання карбомера (карбополу) 934 Р, гідроксіетилцелюлози та натрію альгінату. Для нейтралізації водних дисперсій карбополу використані розчини амоніаку, натрію гідроксиду та триетаноламін. Приготування гелю здійснювалось при кімнатній температурі за двома варіантами технології: шляхом змішування готового гелю і приготовлених розчинів діючих речовин (технологія №1) та шляхом введення діючих речовин до дисперсійного середовища перед загущенням гелю (технологія №2). На підставі комплексу проведених органолептичних, фізико-хімічних і структурно-механічних досліджень були обрані гелеутворювач, нейтралізуючий агент, гідрофільний неводний розчинник та їх оптимальні концентрації у складі препарату. Показано, що термічні ефекти зразків діючих речовин, гелевої основи і гелю «Фіторин-плюс» мають подібний характер, що може суб'єктивно свідчити про відсутність хімічної взаємодії між компонентами препарату і підтверджувати правильність розробленої технології.*

**РАЗРАБОТКА ТЕХНОЛОГИИ НАЗАЛЬНОГО ГЕЛЯ «ФИТОРИН-ПЛЮС» ДЛЯ ЛЕЧЕНИЯ АЛЛЕРГИЧЕСКОГО РИНИТА**

***О.А.Рухмакова, Т.Г.Ярных***

***Ключевые слова:*** *технология; назальный гель; аллергический ринит*

*На современном фармацевтическом рынке среди симптоматических назальных средств имеются преимущественно синтетические лекарственные препараты зарубежного производства. Значительная их часть выпускается в форме капель или спреев, что обусловливает кратковременность действия препаратов. Большое внимание сегодня в лечении аллергического ринита привлекают назальные гели. Цель работы – разработка назального*

*средства на основе веществ растительного происхождения в форме геля под условным названием «Фиторин-плюс». В качестве действующих веществ использованы сухой экстракт корня солодки, эфирные масла сосны и эвкалипта. С целью выбора структурообразующего компонента исследована возможность применения карбомера (карбопола) 934 Р, гидроксиэтилцеллюлозы и натрия альгината. Для нейтрализации водных дисперсий карбопола использованы растворы аммиака, натрия гидроксида и триэтаноламин. Приготовление геля осуществлялось при комнатной температуре по двум вариантам технологии: путем смешивания готового геля и приготовленных растворов действующих веществ (технология №1) и путем введения действующих веществ в дисперсионную среду перед сгущением геля (технология №2). На основании комплекса проведенных органолептических, физико-химических и структурно-механических исследований были выбраны гелеобразователь, нейтрализующий агент, гидрофильный неводный растворитель и их оптимальные концентрации в составе препарата. Показано, что термические эффекты образцов действующих веществ, гелевой основы и геля «Фиторин-плюс» имеют сходный характер, что субъективно свидетельствует об отсутствии химического взаимодействия между компонентами препарата и подтверждает правильность разработанной технологии.*

*Recommended by Doctor of Pharmacy, professor O.A.Ruban*

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# THE CHOICE OF THE RATIONAL TECHNOLOGY OF "DENTATRYHIN" GEL

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*Key words: dentistry; gel; technology research; structural-mechanical properties*

*Particular attention when developing drugs is paid to the choice of the rational technology. The technological process is an important aspect affecting the quality and stability of a drug, and should consist of a system of interconnected and evidence-based operations. The effect of the active components of "Dentatryhin" gel (triclosan, aminocaproic acid and lavender oil) in the concentrations specified on the structural-mechanical and physicochemical properties of test samples of the gels based on Carbomer 934 P has been studied; it is important from the point of view of consumer and technological characteristics of the medicinal product. It has been proven that the combined gel "Dentatryhin" developed for treating gingivitis represents a structured system with moderate thixotropic properties and satisfactory consumer characteristics. The rational technology of a dental gel has been experimentally determined; it consists of several sequential steps such as preparing a gel base, first introducing the alcohol solutions of triclosan and lavender oil into it, and then an aqueous solution of aminocaproic acid with the subsequent homogenization.*

One of the most common causes of periodontitis and tooth loss is gingivitis of different etiology. Distribution of inflammation to the mucous membrane of the alveolar portions in all teeth of the upper and lower jaws is characteristic for the generalized gingivitis process. Therefore, creation of new drugs for topical use with the antimicrobial, anti-inflammatory and hemostatic effect is needed for effective pharmacotherapy of gingivitis [6, 11, 12, 15-8].

The most rational dosage form for topical treatment of gingivitis is gel. Dental gels are very popular because they are easy to apply on the gingival tissue, they are well distributed and absorbed in the mucous membrane of the gingival tissue. This dosage form provides local and uniform release of active substances, creates their high therapeutic concentration in sites of their application without a significant increase of the drug level in the systemic circulation [1, 3, 5, 6].

Using the comprehensive studies the composition of a new drug – "Dentatryhin" gel with triclosan, aminocaproic acid and lavender oil to treat gingivitis has been developed [7-9]. It should be noted that the high therapeutic activity of the drug can be achieved only with the right combination of active components and the base. The composition of the drug should be grounded on the basis of scientific experiments on the choice of active substances and excipients, as well as their desired concentration [4, 5, 10].

One of the most important factors affecting the quality and stability of the gel is also the technology of its production. The technological production process should consist of the rational planned system of interconnected processes, each technological operation must be substantiated.

The aim of our work is to choose and develop the rational technology of preparing a dental gel "Dentatryhin" in the laboratory and industrial conditions.

**Materials and Methods**

The study subjects were the samples of the gels based on Carbomer 934 P produced by "Lubrizol" company. The active substances were introduced in them in the concentrations chosen on the basis of the literature search and grounded due to biological and microbiological research, namely triclosan – 0.5%, aminocaproic acid – 5.0%, lavender oil – 0.5% [1, 3, 7-9, 14]. To develop pilot batches of "Dentatryhin" gel for treating gingivitis the ingredients were introduced under the laboratory conditions of its preparation proposed. The following samples were prepared: Sample No.1 (gel base + triclosan + lavender oil), Sample No.2 (gel base + lavender oil), Sample No.3 (gel base + aminocaproic acid), Sample No.4 ("Dentatryhin" gel with triclosan, aminocaproic acid, lavender oil, nipagin, Carbomer 934 P, sorbitol, sodium hydroxide, ethyl alcohol and purified water in its composition), Sample No.5 (gel base + triclosan).

The standard equipment required in the production of soft dosage forms was used. When developing the technology a MM-1000 mechanical stirrer of "BioSan" company (Latvia) was used. The structural-mechanical studies were performed on a "Brookfield DV-II + PRO" viscometer with a SC 4-21 rotary spindle (USA) at 15-25°C (according to the SPhU) [2]. The rheological parameter – mechanical stability (MS) – was calculated as the ratio of tensile strength to fracture ($\tau 1$) to the value limit after fracture ($\tau 2$). The pH values were determined potentiometrically using a "pH Meter Metrohm 744" device (Germany) [13].

**Results and Discussion**

In order to develop the rational technology the effect of each active ingredient on the structural-mechanical properties of the samples of the gel was studied. For an objective evaluation of the results the study of a

Table 1

Structural-mechanical, physical and chemical characteristics of the test sample of the gels

| Name, indicator | Sample No.1 | Sample No.2 | Sample No.3 | Sample No.4 | Sample No.5 |
|---|---|---|---|---|---|
| Appearance | Semitransparent gel with the odour of lavender essential oil | Semitransparent gel with the odour of lavender essential oil | Transparent gel with little odour of ethanol | Transparent gel with the odour of lavender essential oil | Non-transparent odourless gel |
| pH | 6.22±0.02 | 6.01±0.03 | 6.65±0.01 | 6.55±0.01 | 6.45±0.02 |
| Structural viscosity, η mPa · s at 20 rev/s | 10000 | 13000 | 11300 | 13100 | 13000 |
| Structural viscosity, η mPa · s at 35 rev/s | 6900 | 9000 | 7800 | 9200 | 10000 |
| Structural viscosity, η, mPa · s at 100 rev/s | 3900 | 4600 | 4900 | 4600 | 7850 |
| MS | 1.01 | 1.10 | 1.01 | 1.05 | 1.46 |

Note: n = 5.

number of structural-mechanical and physicochemical parameters was performed (Table 1).

As can be seen from the results of Table 1, all samples of gels under research had a high structural viscosity value at certain speeds of a gate mixer.

The effect of the active substances in certain concentrations on the structural-mechanical properties of the experimental samples of gels was studied. It is important in terms of consumer and technological characteristics. According to the results obtained the rheograms of the samples studied were constructed in the "shear rate – shear stress" coordinates. As can be seen from Fig., all samples of gels had the plastic type of flow, i.e. introduction of active substances and excipients had no effect on the change of the type of flow. Samples No.1-3 had the lowest levels of rheoparameters.

It is known that gel bases based on acrylic acid copolymers exhibit the best structural-mechanical properties at neutral and slightly alkaline pH [4, 10]. Since essential oils have the acidic pH value, Samples No.1 and 2, respectively, show low values of rheoparameters.

The presence of ethanol – the solvent of lavender oil and triclosan (due to the dielectric constant of the solvent) affects decrease of the rheoparameters [10]. As for Sample No.3, it is known that the chemical structure of aminocaproic acid allows it to act as an additional neutralizing agent (the presence of the amino groups). However, in this case, re-neutralization was observed, and it led to unfolding of the polymer macromolecule chains and decrease of the rheoparameters. The rheogram of the gel developed is intermediate between the rheograms of other samples of the gel, suggesting the

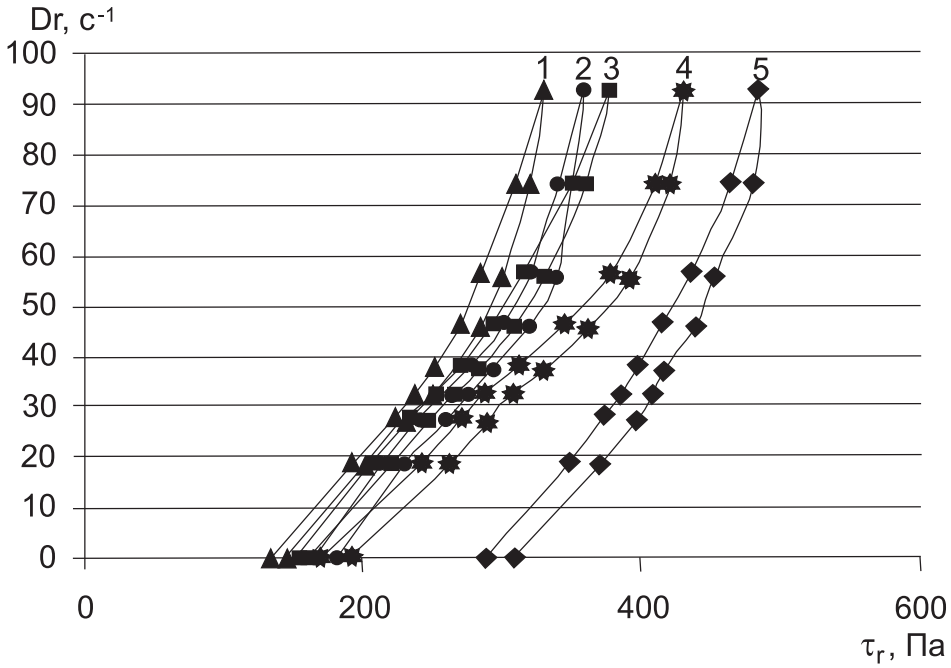


Fig. The rheogram of the samples of gels where: 1 – base + triclosan + lavender oil, 2 – base + lavender oil, 3 – base + aminocaproic acid, 4 – gel for treating gingivitis, 5 – base + triclosan.

Table 2

The study of physical and chemical properties of "Dentatryhin" gel samples prepared by different technologies

| Name | Technology No.1 | Technology No.2 |
|---|---|---|
| Appearance | Homogeneous semitransparent gel-like mass without impurities | Homogeneous semitransparent gel-like mass without impurities |
| pH (10% solution) | 6.55±0.01 | 6.35±0.01 |
| Structural viscosity, mPa · s at 20 rev/min | 13100 | 15000 |
| Structural viscosity, mPa · s at 35 rev/min | 9200 | 10000 |
| Structural viscosity, mPa · s at 100 rev/min | 4600 | 4600 |
| MS | 1.05 | 1.12 |

Note: n = 5.

stability of the sample during storage. For a more complete study of the experimental samples, the MS values characterizing the degree of the structure destruction in the irreversible deformation process were calculated. The data obtained (Table 1) are confirmed by the calculated values of MS.

The samples of "Dentatryhin" gel were prepared at room temperature by two technologies given below.

*Technology No.1.* According to the classical scheme the gel base with Carbomer was prepared [4, 10]. Then at the slow speed of a gate mixer (not more than 70 rev/min) 70% of sorbitol solution was added to the gel base prepared. In parallel, lavender oil and triclosan were dissolved in ethanol, and then the transparent solution obtained was added to the gel base at slow speeds as well. The required number of aminocaproic acid was dissolved in the calculated amount of purified water and injected into the gel. Homogenization of the gel was carried out in a reactor with a gate mixer for 15 minutes while vacuuming to avoid the process of drug aeration.

*Technology No.2.* It differs by the fact that at first an aqueous solution of aminocaproic acid was introduced to the gel base obtained (gelation agent, sorbitol, preservative, purified water), and then the alcohol solution of triclosan and lavender oil. Homogenization of the gel was carried out in a reactor under the same conditions as in technology No.1. The data obtained are presented in Table 2.

As can be seen from Table 2, the sample prepared by technology No.1 has more alkaline pH, providing a more stable gel system; it, in turn, is explained by forming the intermolecular bonds. As a result, a temporary net structure that prevents degradation of the sample is formed.

The mechanical strength of the sample prepared by technology No.2 is much more than in the sample prepared by technology No.1, it can create difficulties in the production process and poor consumer properties.

Thus, on the basis of the experiments conducted and the structural-mechanical properties studied technology No.1 was selected since the sample prepared by it had better structural-mechanical (MS close to perfect – 1.0, better extrusion properties) and technological properties.

CONCLUSIONS

Using the structural-mechanical and physicochemical research the effect of active substances of the gel (triclosan, aminocaproic acid and lavender oil) on stability of the gel base with Carbomer 934 P has been studied.

The rational technology of a dental "Dentatryhin" gel has been selected; it comprises the following steps such as preparing a gel base; introduction of alcohol solutions of triclosan and lavender oil; introduction of the aqueous solution of aminocaproic acid to the gel base; homogenization of the gel; dispensing and packing of the gel.

REFERENCES


1. *Безпала Ю.О. Перспективи використання карбомерів у технології гелів для лікування запальних захворювань слизової оболонки рота // Товарознавчі аспекти споживчих товарів: матер. II наук.-практ. конф. за міжнар. участю, м. Харків, 19 березня 2013 р. – Х.: НФаУ, 2013. – С. 71.*
2. *Державна фармакопея України / Державне підприємствово «Науково-експертний фармакопейний центр». – 1-е вид., 1 доп. – Х.: РІРЕГ, 2004. – 520 с.*
3. *Допоміжні речовини в технології ліків: вплив на технологічні, споживчі, економічні характеристики і терапевтичну ефективність: навч. посіб. для студ. вищ. фармац. навч. закл. / Авт.-уклад.: І.М.Перцев, Д.І.Дмитрієвський, В.Д.Рибачук та ін.; за ред. І.М.Перцева. – Х.: Золоті сторінки, 2010. – 600 с.*
4. *Козир Г.Р., Тихонов О.І. // Фармац. журн. – 2003. – №1. – С. 78-82.*
5. *Лебединець О.В., Баранова І.І., Петровська Л.С. // Укр. журн. клін. та лабораторної медицини. – 2010. – Т. 5, №3. – С. 57-60.*
6. *Мюллер Х.П. Пародонтология. – Львов: Изд-во «ГалДент», 2004. – 256 с.*
7. *Портянко В.С., Коваленко Св.М., Малоштан Л.М. // Фармац. журн. – 2014. – №3. – С. 83-86.*

8. *Портянко В.С., Коваленко Св.М., Стрілець О.П. // Проблеми екологічної та медичної генетики і клінічної імунології. – 2012. – Вип. 5 (113). – С. 480-484.*
9. *Портянко В.С. Розробка складу гелю комплексної дії для лікування гінгівіту // Матер. Всеукр. наук. конф. студентів та молодих учених «Актуальні питання створення нових лікарських засобів», м. Харків, 20-21 квітня 2011 р. – Х.: НФаУ, 2011. – С. 221-221.*
10. *Ролік С.М., Пімінов О.Ф, Лукієнко О.В. // Укр. журн. клін. та лабораторної медицини. – 2009. – Т. 4, №1. – С. 12-15.*
11. *Цепов Л.М. Заболевания пародонта: взгляд на проблему. – М.: МЕДпресс-информ, 2006. – С. 8-70.*
12. *Genco R. // J. Periodontol. – 2008. – Vol. 79, №8. – P. 1609-1611.*
13. *Malkin A.Ya. Rheology Concepts, Methods, and Applications. – London: Applied Science Publishers, 2006. – 474 p.*
14. *Muramatsu M., Kanada K., Nishida A. et al. // Intern. J. of Pharmac. – 2000. – Vol. 199, №1. – P. 77-83.*
15. *Offenbacher S., Barros S.P., Beck J.D. // J. Periodontol. – 2008. – Vol. 79, №8. – P. 1577-1584.*
16. *Picout D.R., Ross-Murphy S.B. // The Sci. World J. – 2003. – Vol. 24, №3. – P. 105-121.*
17. *Van Dyke T.E. // J. Periodontol. – 2008. – Vol. 79, №8. – P. 1601-1608.*
18. *Van Dyke T.E., Winkelhoff A.J. // J. Clin. Periodontol. – 2013. – Vol. 40, №14. – P. 1-7.*

**ВИБІР РАЦІОНАЛЬНОЇ ТЕХНОЛОГІЇ ГЕЛЮ «ДЕНТАТРИГІН»**

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***Ключові слова:*** *стоматологія; гель; технологічні дослідження; структурно-механічні властивості*

*Особлива увага при розробці лікарських препаратів приділяється вибору раціональної технології. Технологічний процес – це важливий аспект, що впливає на якість і стабільність лікарського засобу, який повинен складатися з системи взаємопов'язаних та науково-обгрунтованих операцій. Вивчено вплив активних компонентів гелю «Дентатригін» (триклозану, амінокапронової кислоти і лавандової олії) у встановлених концентраціях на структурно-механічні та фізико-хімічні властивості експериментальних зразків гелів на основі карбомера марки 934 Р, що є важливим з точки зору споживчих і технологічних характеристик лікарського засобу. Доведено, що розроблений комбінований гель «Дентатригін» для лікування гінгівітів являє собою структуровану систему з помірними тиксотропними властивостями та задовільними споживчими характеристиками. Експериментально встановлена раціональна технологія стоматологічного гелю, яка складається з декількох послідовних операцій: приготування гелевої основи, введення в неї спочатку спиртових розчинів триклозану і лавандової олії, а потім водного розчину амінокапронової кислоти з подальшою гомогенізацією.*

**ВЫБОР РАЦИОНАЛЬНОЙ ТЕХНОЛОГИИ ГЕЛЯ «ДЕНТАТРИГИН»**

***В.С.Кучеренко, Св.Н.Коваленко, И.И.Баранова***

***Ключевые слова:*** *стоматология; гель; технологические исследования; структурно-механические свойства*

*Особое внимание при разработке лекарственных препаратов уделяется выбору рациональной технологии. Технологический процесс – это важный аспект, влияющий на качество и стабильность лекарственного средства, который должен состоять из системы взаимосвязанных и научно-обоснованных операций. Изучено влияние активных компонентов геля «Дентатригин» (триклозана, аминокапроновой кислоты и лавандового масла) в установленных концентрациях на структурно-механические и физико-химические свойства экспериментальных образцов гелей на основе карбомера марки 934 Р, что является важным с точки зрения потребительских и технологических характеристик лекарственного средства. Доказано, что разработанный комбинированный гель «Дентатригин» для лечения гингивитов представляет собой структурированную систему с умеренными тиксотропными свойствами и удовлетворительными потребительскими характеристиками. Экспериментально установлена рациональная технология стоматологического геля, которая состоит из нескольких последовательных операций: приготовления гелевой основы, введения в нее сначала спиртовых растворов триклозана и лавандового масла, а затем водного раствора аминокапроновой кислоты с последующей гомогенизацией.*

*Recommended by Doctor of Pharmacy, professor N.P.Polovko*

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# THE STUDY OF THE FOAMING ABILITY OF SOME SURFACTANTS

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National Medical Academy of Postgraduate Education named after P.L. Shupyk

*Key words: surfactants; foaming ability; stability and height of the foam*

*The foaming ability of some surfactants, namely such parameters as the height of the foam column and foam stability, has been studied. It has been found that in solutions with one surfactant there is no specific dependence between the foam height and foam stability. It has been proven experimentally that the active phase of foaming (the height of the foam column) lasts up to 100-150 s. Starting from 150 s the height of the foam decreases, and this process lasts up to 300 s. It has been shown that despite the fact that the surfactants studied form foam in solutions, there is no possibility to select the optimal surfactant. According to the studies conducted it is due to the fact that a stable foam is formed by combining two or more surfactants. The data obtained show that the test solutions are not suitable for use in the drug composition because it is impossible to guarantee the quality of the foam. Therefore, to ensure the quality of foam-forming composition it is rational to continue further studies of the combination of two or more surfactants in solutions.*

Natural and synthetic surfactants are an essential component in the foaming compositions. The ability to absorption at the interphase boundary and aggregation in the bulk solution is significant for each surfactant [6, 7]. Adsorbing at interfaces and forming micelles the surfactants contribute to dissolution, emulsification, foaming and some other processes that occur in solutions [4].

Along with the surface activity of the surfactant itself, the foaming ability of a surfactant is characterized by the mechanical strength and viscosity of the films formed [2, 4, 6].

Methods for determining the ability of the foaming ability can be divided into static and dynamic although this division is rather conventional; and taking into account the equipment used in the studies in some cases there is no difference between these methods [5, 6]. Determination by dynamic methods is carried out under continuous mechanical action on the solution to prevent the possibility of running off the foam from it [3, 5]. Under dynamic conditions the volume of the foam measured is determined by the ratio between the rate of its formation and its destruction, and the volume of foam under static conditions is dependent on the speed of bubbles dissolution. The foaming ability of surfactant solutions is the characteristic that must be considered when developing foam formulations. This is due to the fact that formation of a stable foam is a guarantee of quality of the foam formulation.

The aim of this study was to investigate the foaming ability of some surfactants determining such indicators as the height of the foam column and foam stability.

**Materials that Methods**

The foaming ability of the surfactant solutions was determined on a Ross-Miles device at 50±2°C according to the GOST 22567.1-77 [1].

The objects of the study were such surfactants as sodium lauryl sulfate, emulsifier No.1, OC 20 (macrogol cetostearyl ester), polysorbate 80, sorbitan laurate, sodium docusate, PEG 6 stearate (glycol stearate), sucrose palmitate, PEG 75 stearate (cetyl alcohol (and) glyceryl stearate), cocamidopropylbetaine, PEG 100 stearate (glyceryl stearate and PEG 100 stearate), glyceryl laurate, distilled monoglycerides, wax emulsion, dodecyl dipropylene triamine, polysorbate 20, dodecyl-dimethyl ammonium chloride, polyhexamethyleneguanidine, miramistin.

Test solutions were prepared with the surfactant concentration of 7%.

**Results and Discussion**

Fig. 1 (A, B, C, D) presents the results of our studies in the form of a graphical dependence of the height of the foam column on time (300 s).

A comparative analysis of the data in Fig. 1 shows that the indicator of the height of the foam column of such surfactants as PEG 6 stearate, PEG 75 stearate, PEG 100 stearate, wax emulsion, emulsifier No.1, distilled monoglycerides, sucrose palmitate and glyceryl laurate is inferior to the height of the foam column of such surfactants as sodium lauryl sulfate, OC 20, polysorbate 80, sorbitan laurate, sodium docusate, cocamidopropylbetaine, dodecyl dipropylene triamine, polysorbate 20, dodecyl-dimethyl ammonium chloride, polyhexamethyleneguanidine and miramistin. The active phase of foaming (the height of the foam column) lasts up to 100-150 s. Starting from 150 s the height of the foam decreases, and this process lasts up to 300 s.

When developing a drug the indicator of the foam stability is also an important characteristic because, in our opinion, it directly affects the therapeutic activity of the drug. Fig. 2 shows the results of the study of the foam stability of the surfactants mentioned above.

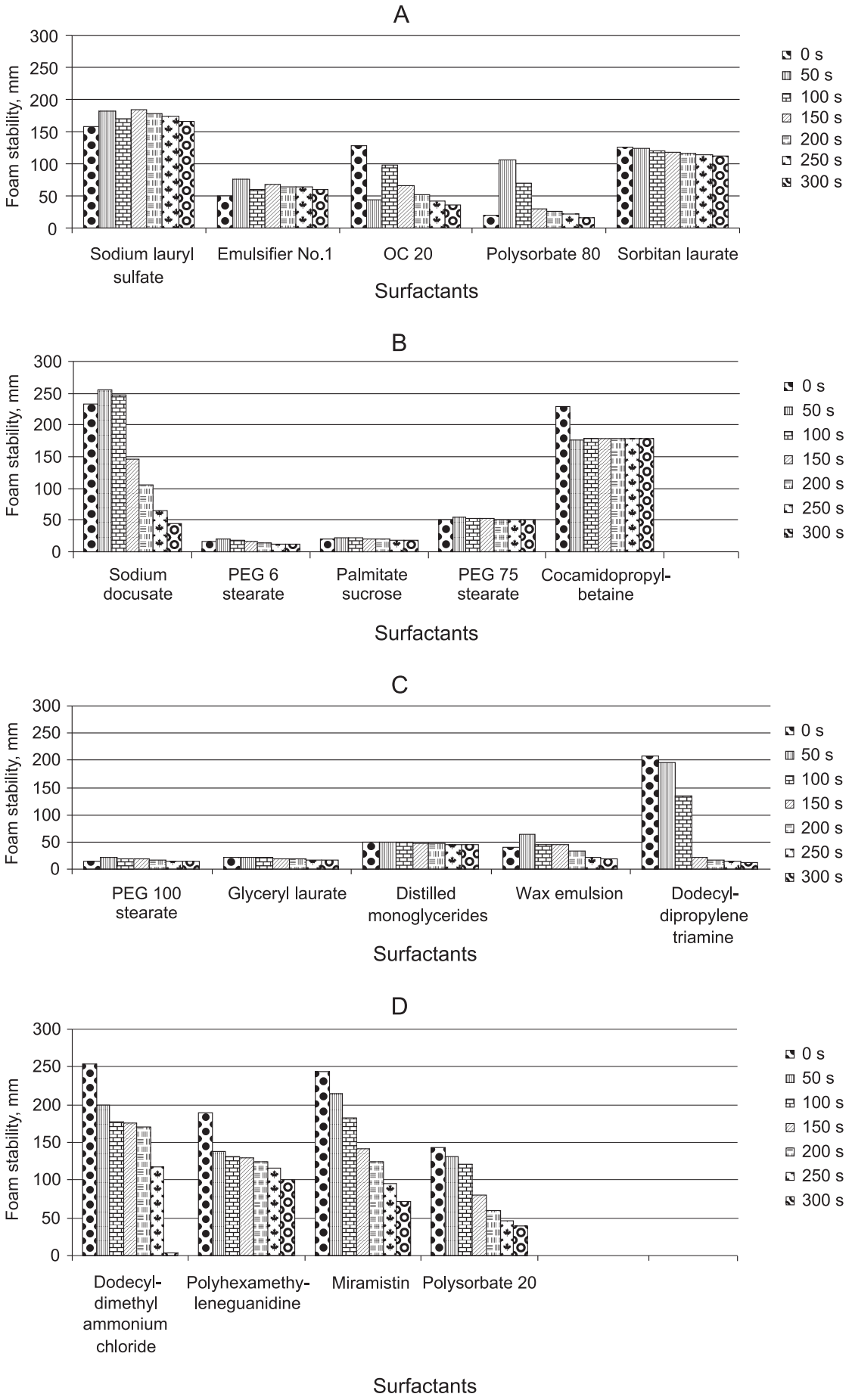


Fig. 1. The foaming ability of surfactants depending on time.

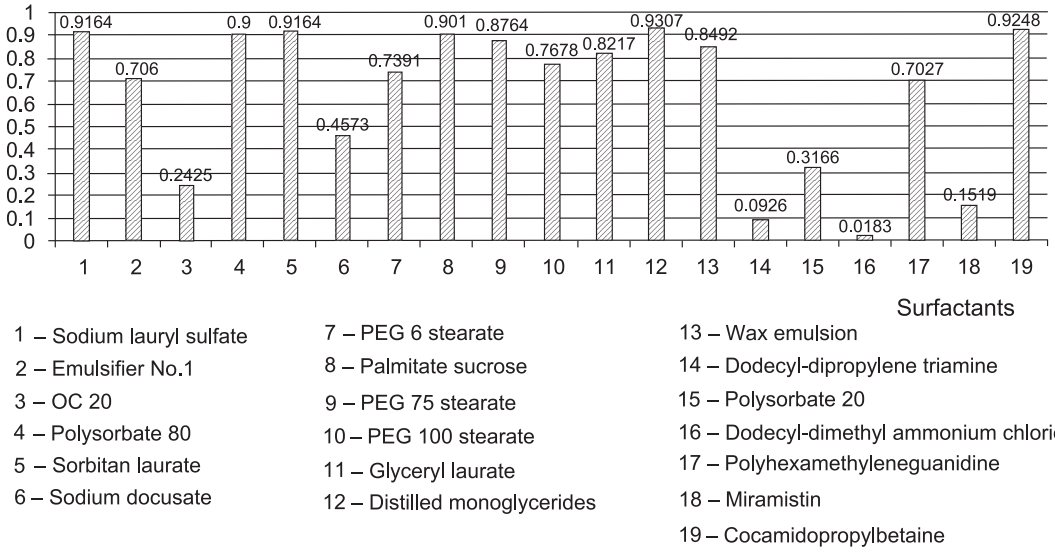


Fig. 2. Stability of the surfactant foam.

It has been found experimentally that sodium lauryl sulfate, emulsifier No.1, polysorbate 80, sorbitan laurate, PEG 6 stearate, sucrose palmitate, PEG 75 stearate, cocamidopropylbetaine, PEG 100 stearate, glyceryl laurate, distilled monoglycerides, wax emulsion and polyhexamethyleneguanidine form a stable foam. It should be noted that there is no direct relationship between the height of the foam and its stability.

## CONCLUSIONS

The data obtained show that despite the fact that the surfactants studied form foam in solutions, there is no possibility to select the optimal surfactant. According to the studies previously conducted a stable foam is formed by combining two or more surfactants; therefore, foam-forming properties of mixtures of hydrophilic and hydrophobic surfactants will be studied in future.

## REFERENCES

1. *Средства моющие синтетические. Метод определения пенообразующей способности: ГОСТ 22567.1-77 (СТ СЭВ 4155-83). – [Взамен ГОСТ 22567.1-77]. – Введ. 01.05.86. – М.: Изд-во стандартов, 1986. – С. 1-6.*
2. *Холмберг К., Йёнссон Б., Кронберг Б., Линдман Б. Поверхностно-активные вещества и полимеры в водных растворах. – М.: БИНОМ. Лаборатория знаний, 2007. – 528 с.*
3. *Langevin D. // Adv. Colloid Interface Sci. – 2000. – Vol. 88. – P. 209-222.*
4. *Lunkenheimer K., Malysa K., Winsel K. et al. // Langmuir. – 2010. – Vol. 26. – P. 3883-3888.*
5. *Russev S.C., Alexandrov N., Marinova K.G. // Rev. Sci. Instr. – 2008. – Vol. 79. – P. 102-104.*
6. *Stubenrauch C., Fainerman V.B. // J. Phys. Chem. B. – 2005. – Vol. 109. – P. 1505-1509.*
7. *Stubenrauch C., Khristov Kh. // J. Colloid Interface Sci. – 2005. – Vol. 286. – P. 710-718.*

**ДОСЛІДЖЕННЯ ПІНОУТВОРЮЮЧОЇ ЗДАТНОСТІ ДЕЯКИХ ПОВЕРХНЕВО-АКТИВНИХ РЕЧОВИН**

***А.О.Дроздова***

***Ключові слова:*** *поверхнево-активні речовини; піноутворююча здатність; стійкість і висота піни*

*Вивчена піноутворююча здатність деяких поверхнево-активних речовин, а саме такі їх показники як висота стовпа та стійкість піни. Встановлено, що в розчинах, які містять одну поверхнево-активну речовину, не існує певної залежності між висотою піни і її стійкістю. Експериментально доведено, що активна фаза піноутворення (висота стовпа піни) триває 100-150 с. Починаючи з 150 с, висота стовпа піни зменшується, а даний процес продовжується до 300 с. Показано, що досліджувані поверхнево-активні речовини в розчинах хоча і утворюють піну, але обрати оптимальну поверхнево-активну речовину неможливо. Це пов'язано з тим, що стійка піна, згідно з проведеними дослідженнями, утворюється при поєднанні двох і більше поверхнево-активних речовин. Отримані дані свідчать про те, що досліджувані розчини не придатні для використання в складі препарату, так як неможливо гарантувати якість піни. Тому доцільно подальше вивчення сумішей двох і більше поверхнево-активних речовин у розчинах для гарантованої якості піноутворюючого складу.*

**ИССЛЕДОВАНИЕ ПЕНООБРАЗУЮЩЕЙ СПОСОБНОСТИ НЕКОТОРЫХ ПОВЕРХНОСТНО-АКТИВНЫХ ВЕЩЕСТВ**

***А.А.Дроздова***

***Ключевые слова:*** *поверхностно-активные вещества; пенообразующая способность; устойчивость и высота пены*

*Изучена пенообразующая способность некоторых поверхностно-активных веществ, а именно такие показатели как высота столба пены и устойчивость пены. Установлено, что в растворах с одним поверхностно-активным веществом нет зависимости между высотой пены и ее устойчивостью. Экспериментально доказано, что активная фаза пенообразования (высота столба пены) продолжается до 100-150 с. Начиная с 150 с, высота столба пены уменьшается, а данный процесс длится до 300 с. Показано, что исследуемые поверхностно-активные вещества в растворах хотя и образуют пену, но выбрать оптимальное поверхностно-активное вещество не представляется возможным. Это связано с тем, что стойкая пена, согласно проведенным исследованиям, образуется при сочетании двух и больше поверхностно-активных веществ. Полученные данные свидетельствует о том, что исследуемые растворы не пригодны для использования в составе препарата, так как невозможно гарантировать качество пены. Поэтому целесообразно дальнейшее изучение сочетания двух и более поверхностно-активных веществ в растворах для гарантированного качества пенообразующего состава.*

# ОРГАНІЗАЦІЯ ТА ЕКОНОМІКА ФАРМАЦІЇ

*Recommended by Doctor of Pharmacy, professor A.S.Nemchenko*

UDC 65.011.47:336.146:640.522.2

## ASSESSMENT OF THE RATIONAL USE OF BUDGETARY FUNDS ON DRUG SUPPLY BY HEALTHCARE INSTITUTIONS UNDER THE CONDITIONS OF INTRODUCTION OF THE FORMULARY SYSTEM

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*Key words: formulary system; local formulary; drugs; ABC/VEN-analysis; index of utility; pharmacotherapeutic commission*

*The growth of expenses on drug supply in healthcare requires effective methods for their analysis in the specific healthcare institutions. On the example of a conditional healthcare institution an integrated ABC/VEN-analysis of the expenses on the drug supply for the year has been conducted using the index of utility for each drug. The results of the analysis performed will allow to optimize the purchase of drugs in accordance with the structure of diseases registered in the given healthcare institution, as well as the structure of the local formulary.*

One of the main tasks of healthcare of any country is providing medical care of the adequate quality. According to experts of the World Health Organization, any country in the world does not have sufficient resources to healthcare. Even countries with high economic growth have to look for mechanisms to optimize the use of health budgets. In recent years particularly acute this problem arose in Ukraine.

The experience of a number of economically developed countries allows to note a significant role, which can be played by introduction of the formulary system based on the standards of treatment and the principles of evidence-based medicine in solving these problems. Introduction of the formulary system is the most effective means of economical use of budgetary funds allocated for the drug supply [3, 10, 11, 13, 14].

An important element of the quality management system providing availability, effectiveness and safety of drugs is conducting the ABC/VEN-analysis by healthcare institutions on using budgetary funds for the drug supply. To implement this task according to the requirements of the order of the Ministry of Public Health of Ukraine dated 22. 07. 2009 No. 529 "About creating a formulary system for the drug supply of healthcare institutions" the pharmacotherapeutic commission (PTC) is created by the order of the chief physician of a healthcare institution. An important task of such commission together with development and constant updating of the local formulary of drugs is providing physicians with information on the rational pharmacotherapy, as well as conducting the ABC-, VEN-analysis on using drugs, determination of rationality of their purchase in accordance with the priorities of pharmacotherapy of diseases registered in healthcare institutions (statistical data form No.20) [4, 5, 14].

Conducting of the clinical and economic analysis is the most time consuming chain in the process of introduction of the formulary system. Despite the obvious perspective and the rapid development of this direction of research, its effectiveness is still rather low. One of the causes admitted by researchers is the lack of awareness of physicians concerning the issues of pharmacoeconomics and conservatism in prescribing drugs that is probably associated with this. Existing practical methods and guidelines are largely limited. For example, the ABC/VEN-analysis is mostly conducted by researchers while studying the system of using drugs for a certain nosological form of the disease, or separate groups of drugs, and it makes its implementation in practical work of healthcare institutions is quite time-consuming and insufficiently informative. When forming the local formulary there is virtually no experience in assessing the drug utility that would allow PTC to estimate objectively the degree of provision of patients with drugs, as well as the correspondence of the financial costs to the structure of diseases registered in the given healthcare institution [1, 2, 3, 8, 9].

The lack of specialists with the knowledge in the fields of economics, healthcare and pharmacy in most healthcare institutions, as well as computer technologies determine the need of development of the corresponding methodological recommendations [5, 6].

With introduction of the formulary system, in particular, creation of local formularies of drugs in healthcare institutions, and necessity of regular conducting of the ABC/VEN-analysis, the members of PTC must

master the method of its conducting with the use of computer technologies.

Based on the above the aim of our study was to practice the method of the integrated ABC/VEN-analysis of using drugs when forming and updating local formularies, as well as determination of rationality of their purchase and the correspondence to the structure of the patients admitted to hospital.

**Materials and Methods**

For the retrospective assessment of the rational use of the budgetary funds of a certain healthcare institution to purchase drugs such method of clinico-economic analysis as the ABC/VEN-analysis is used. Combination of the ABC and VEN analysis gives an idea about what drugs that are vital (V), essential (E); nonessential (N) take the most significant place in the structure of expenses of the healthcare institution for the drug supply.

The ABC-analysis (the Pareto principle) is a standard method used in the world when assessing current expenditures for purchase of drugs and their planning for the next year under the conditions of introduction of the formulary system.

The current modification of the VEN-analysis is assessment of the drug utility, i.e. degree of the need to use the given drug for pharmacotherapy of a specific disease, and it is extremely important, in particular, when deciding the reasonability of including the corresponding drug to the local formulary of the healthcare institution [1, 7].

In this case it is possible to determine the level of utility in the form of specific indexes VEN (UV, UE, UN) adding the code of a disease to them according to ICD-10. For example, according to the standardized protocol of medical care approved by the order of the Ministry of Public Health of Ukraine dated 19.03.07 No. 128 "On approval of clinical protocols of medical care in the specialty "Pulmonology" in drug therapy of community-acquired pneumonia (ICD-10 code – J15.9) the solution for infusion "Leflocin" will have the index of utility UV J15.9. This index can be further objectify if the level of efficiency and safety of drugs is added to it on the scale of levels of evidence. The index "0" refers to the utility of the drug, which is absent in clinical protocols of medical care of diseases registered in the given healthcare institution in the current State formulary of drugs, or in the List of Drugs that can be purchased by healthcare institutions fully or partially financed from the budget [12].

The benefits of implementing such analysis is obvious since it closely links the structure of morbidity registered in the healthcare institution (ICD-10 code), the corresponding protocols of medical care and drugs used in the drug therapy.

By definition, a local formulary should contain as many drugs as possible, which in relation to diseases registered in the particular healthcare institution must be referred to vital and essential drugs, and, therefore, have the index of utility ($U_V$) and ($U_E$), and only in extreme cases – to nonessential drugs ($U_N$).

It should be noted that at the beginning of the study (2012) the most common approach to development of the local formulary was as follows: all drugs used at that moment in the healthcare institution were considered a basic formulary, which in the future should gradually be updated and improved according to the principles of the formulary system. That is, based on the results obtained, PTC of the healthcare institution planned to develop pharmacoeconomically reasonable local formularies that would have to meet the requirements of normative legal documents. An important role in this process was given to the ABC/VEN-analysis [11].

In our research 398 healthcare institutions of the secondary and tertiary levels from the Dnipropetrovsk, Vinnytsia, Kharkiv, Kyiv and other regions of Ukraine took part, they developed their local formularies (LF) for 2013 based on the current of the fifth edition of the State formulary (SF). In order to update and improve LF for 2014 the ABC/VEN-analysis of the rational use of budgetary funds for the purchase of drugs was conducted upon completion of the period under review together with PTC of these healthcare institutions, and the appropriate recommendations concerning improvement of the structure of their purchase, as well as optimization of the content of LF were given.

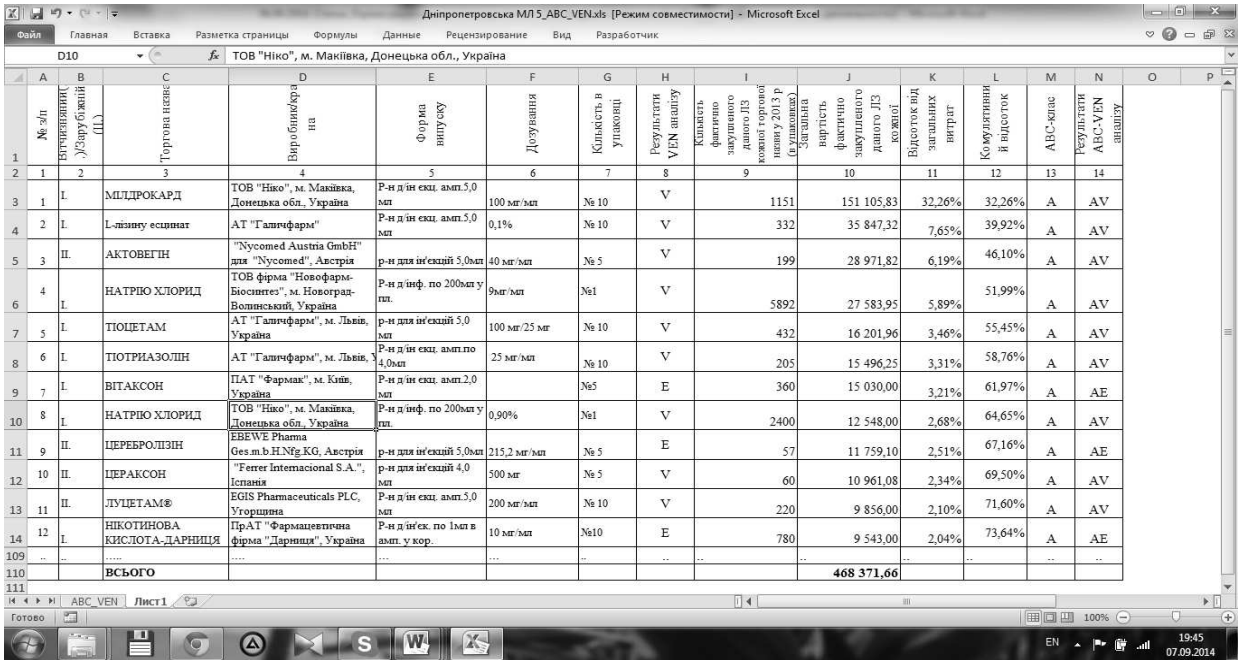
To improve clarity of the process of conducting the integrated ABC/VEN-analysis with the use of computer technologies we consider it expedient to comment it on the example of one of the healthcare institutions in the Dnipropetrovsk region. The basic data are that in 2013 the healthcare institution spent the budgetary funds in the amount of 468371,66 UAH for purchasing 106 names of drugs (data from the local formulary and accounting documents).

The structure of diseases registered in the healthcare institution (statistical data form No.20 for 2013) according to ICD-10 codes is as follows: A00, A08, A04.9, G45, G90-99, I10-13, I20-25, I60, I63, I69, I69.4, I70.8, I177.6, I178, J-00-99, J00-J06, J15.9, J18, J45, K00-K93, K26, K29, K30, K35.9, K 80, K81, K82, K85, K86, L00-L08, L50, M16-19, M42, M54, N 20, N30, R52.0, T36-50, T51, T79.4, T-98, etc.

All trade names of drugs purchased by the healthcare institution within a year were introduced indicating their amount and cost of purchase to the computer in a spreadsheet format with the help of Microsoft Office Excel programme. From the local formulary the data regarding the category of vital importance – V, E, N assigned to each trade name of the drug purchased for the budgetary funds were introduced (Fig.).

With the help of spreadsheet functions drugs were ranked in descending order of the cost for their purchase, the total cost of drugs actually purchased in 2013 was calculated, the expense percentage of the healthcare institution per each individual drug from the sum of the total cost for drug purchase was calculated, the so-called "cumulative percentage" was determined for each drug.

It was further determined which class of ABC should include each of the drugs purchased for the budgetary funds. Drugs, which cumulative percentage was ≤80%, were referred to category A; drugs with the cumulative percentage of 80%≥95% were referred to category B; and drugs with the cumulative percentage of 95%≥100% fell in the category C. Class ABC for each drug was determined using spreadsheets.




| | A | B | C | D | E | F | G | H | I | J | K | L | M | N | O | P |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | № з/п | Вітчизняний /Зарубіжний (ІЛ) | Торгова назва | Виробник/країна | Форма випуску | Дозування | Кількість в упаковці | Результати VEN аналізу | Кількість фактично закупленого даного ЛЗ кожної торгової назви у 2013 р (упаковок) | Загальна вартість фактично закупленого даного ЛЗ кожної | Відсоток від загальних витрат | Кумулятивний відсоток | АВС-клас | Результати АВС-VEN аналізу | | |
| 2 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | | |
| 3 | 1 | I. | МЛДРОКАРД | ТОВ "Ніко", м. Макіївка, Донецька обл., Україна | Р-н д/ін екц. амп.5,0 мл | 100 мг/мл | № 10 | V | 1151 | 151 105,83 | 32,26% | 32,26% | A | AV | | |
| 4 | 2 | I. | L-лізину есцинат | АТ "Галичфарм" | Р-н д/ін екц. амп.5,0 мл | 0,1% | № 10 | V | 332 | 35 847,32 | 7,65% | 39,92% | A | AV | | |
| 5 | 3 | II. | АКТОВЕГІН | "Nycomed Austria GmbH" для "Nycomed", Австрія | р-н для ін'єкцій 5,0мл | 40 мг/мл | № 5 | V | 199 | 28 971,82 | 6,19% | 46,10% | A | AV | | |
| 6 | 4 | I. | НАТРІЮ ХЛОРИД | ТОВ фірма "Новофарм-Біосинтез", м. Новоград-Волинський, Україна | Р-н д/інф. по 200мл у пл. | 9мг/мл | №1 | V | 5892 | 27 583,95 | 5,89% | 51,99% | A | AV | | |
| 7 | 5 | I. | ТІОЦЕТАМ | АТ "Галичфарм", м. Львів, Україна | р-н для ін'єкцій 5,0 мл | 100 мг/25 мг | № 10 | V | 432 | 16 201,96 | 3,46% | 55,45% | A | AV | | |
| 8 | 6 | I. | ТІОТРИАЗОЛІН | АТ "Галичфарм", м. Львів, У | Р-н д/ін екц. амп.по 4,0мл | 25 мг/мл | № 10 | V | 205 | 15 496,25 | 3,31% | 58,76% | A | AV | | |
| 9 | 7 | I. | ВІТАКСОН | ПАТ "Фармак", м. Київ, Україна | Р-н д/ін екц. амп.2,0 мл | | №5 | E | 360 | 15 030,00 | 3,21% | 61,97% | A | AE | | |
| 10 | 8 | I. | НАТРІЮ ХЛОРИД | ТОВ "Ніко", м. Макіївка, Донецька обл., Україна | Р-н д/інф. по 200мл у пл. | 0,90% | №1 | V | 2400 | 12 548,00 | 2,68% | 64,65% | A | AV | | |
| 11 | 9 | II. | ЦЕРЕБРОЛІЗІН | EBEWE Pharma Ges.m.b.H.Nfg.KG, Австрія | р-н для ін'єкцій 5,0мл | 215,2 мг/мл | № 5 | E | 57 | 11 759,10 | 2,51% | 67,16% | A | AE | | |
| 12 | 10 | II. | ЦЕРАКСОН | "Ferrer Internacional S.A.", Іспанія | р-н для ін'єкцій 4,0 мл | 500 мг | № 5 | V | 60 | 10 961,08 | 2,34% | 69,50% | A | AV | | |
| 13 | 11 | II. | ЛУЦЕТАМ® | EGIS Pharmaceuticals PLC, Угорщина | Р-н д/ін екц. амп.5,0 мл | 200 мг/мл | № 10 | V | 220 | 9 856,00 | 2,10% | 71,60% | A | AV | | |
| 14 | 12 | I. | НІКОТИНОВА КИСЛОТА-ДАРНИЦЯ | ПрАТ "Фармацевтична фірма "Дарниця", Україна | Р-н д/ін'єк. по 1мл в амп. у кор. | 10 мг/мл | №10 | E | 780 | 9 543,00 | 2,04% | 73,64% | A | AE | | |
| 109 | .. | .. | ..... | .... | ... | ... | .. | .. | .. | .. | .. | .. | .. | .. | | |
| 110 | | | ВСЬОГО | | | | | | | 468 371,66 | | | | | | |
| 111 | | | | | | | | | | | | | | | | |

Fig. The screenshot of the table with the results of ABC/VEN-analysis for all trade names of the drugs purchased by the healthcare institution for budgetary funds.

With the help of spreadsheet functions the results of the ABC/VEN-analysis for all trade names of drugs were obtained (column 14).

**Results and Discussion**

The results of the ABC/VEN-analysis conducted were listed in Tab. 1 for clarity.

Then for the first time in the practice of the formulary system introduction the index of utility for the drugs purchased was determined with addition of the appropriate codes of diseases in each drug trade name registered in healthcare institutions in according to ICD-10. Ideally, certain diseases codes must correspond to all drugs used in pharmacotherapy of diseases registered in healthcare institutions and included into the local formulary.

The results of the ABC/VEN-analysis with the index of utility determined for each drug purchased for the budgetary funds were listed in Tab. 2.

According to the results of the integrated ABC/VEN-analysis with determination of the level of utility of each drug the following conclusions and recommendations were made by PTC of the healthcare institution:

- Of 106 names of drugs purchased by the healthcare institution, 16 names of drugs, such as mildrocard, l-lysine aescinat, actovegin, etc., are absent in the 5-th edition of the State formulary, and therefore, it is inexpedient to classify them as vital (V) and essential (E) drugs. The share of such drugs was 68.66% of the total expenditures;
- The approaches to the use of drugs of the vital (V) category, which should be given priority when purchasing, should be revised. 83 Drugs were referred to this category, their share in the total budget expenditures was 86.03%. Among them 12 names of drugs (61.20%) are absent in the current State formulary; it is no reason to assign 29 names of drugs (9.44%), such as pentoxifylline, sermion, mucolvan, asparcam, etc., to the category (V).
- The approaches to the use of drugs of the essential drug category (E) should be revised. 22 Drugs were referred to this category, their share was 13.95%. Among them 4 names of drugs (7.46%) are absent in the current State formulary, 8 names of drugs (1.7%), such as ethyl, barboval, ascorbic acid, etc., are not advisable to classify as (E).
- The question concerning increase of the level of the drug supply of the patients with diseases registered in healthcare institutions (statistical data form No. 20

Table 1

Distribution of drugs by the categories of vital importance and their place in the structure of expenses of budgetary funds on drug supply

| In 2013 | | | The share of drugs of each category V; E; N, % | | |
|---|---|---|---|---|---|
| | | | V | E | N |
| | | | 86.03% | 13.96% | 0.01% |
| The share of drugs ranged in classes A, B, C, % | A | 80.09% | AV – 70.60% | AE – 9.49% | AN – 0.00% |
| | B | 15.06% | BV – 11.59% | BE – 3.47% | BN – 0.00% |
| | C | 4.84% | CV – 3.84% | CE – 0.99% | CN – 0.01% |

Table 2

Generalized results of the integrated ABC/VEN-analysis with the index of utility for each drug

| No. | Trade name of the drug, how supplied, dose, package amount | Index of utility of a drug, U | The percentage of total expenditure, % | Conclusions |
|---|---|---|---|---|
| Group (AV) according to the results of the ABC/VEN-analysis | | | | |
| 1 | Mildrocard, sol. f/inj., 5.0 ml amp., 100 mg/ml, No.10 | $U_V$ I20-I25; $U_V$ I60-I69 | 32.26% | Absent in the SPhU, the 5-th ed., therefore it should have the index of utility "0", review the feasibility of using |
| 2 | L-Lysine Aescinat, sol. f/inj., 5.0 ml amp., 0.001, No.10 | $U_V$ I88; $U_V$ I70.8; $U_V$ I77.6 | 7.65% | Absent in the SPhU, the 5-th ed., therefore it should have the index of utility "0", review the feasibility of using |
| 3 | Actovegin, sol. f/inj., 5.0 ml amp., 40 mg/ml, No.5 | $U_V$ I10-13, $U_V$ G45, $U_V$ G90-99 | 6.19% | Absent in the SPhU, the 5-th ed., therefore it should have the index of utility "0", review the feasibility of using |
| 4 | Sodium chloride, sol. f/inj., 200 ml in vial, 9 mg/ml, No.1 | $U_V$ A0.8, $U_V$ T79.4 | 5.89% | No comments |
| | …… | | | |
| | Total of the group (AV) | | 70.60% | |
| and so on | | | | |

for 2013) should be considered according to ICD-10 codes: J15.9, J18, K29, K30, N30, etc.

Because of publication of the sixth edition of the State formulary approved by the order of the Ministry of Public Health of Ukraine dated 08.04.2014 No.252 "On approval of the sixth edition of the State formulary of drugs and providing its availability", and considering the comments received regarding the structure of the drug purchase in 2013, the PTC of healthcare institutions participating in the research made the appropriate changes to the plan of purchasing drugs for budgetary funds for 2014, as well as revised the content of the current local formularies with the purpose of correcting the LF project and the plan of the drug purchase for 2015. The results of the integrated ABC/VEN-analysis of planned indicators of the drug purchase in 2015 indicate qualitative positive changes in the structure of purchasing of categories (V) and (E), as well as in assessing the utility of drugs included to the local formulary.

Summarizing the results of the integrated ABC/VEN-analysis conducted by us with the participation of PTC of healthcare institutions according to the results of drug purchasing in 2013 a number of disadvantages that are common practically for all healthcare institutions may be noted in the process of the local formularies updating and purchasing drugs for budgetary funds.

Obvious signs of irrational use of budgetary funds are as follows:

- the presence of drugs categorized to N (AN) in class A, but they should not be in this class;
- insufficient presence of drugs of V (AV) category in class A, which should be used for treating diseases with a significant specific weight in the structure of diseases registered in healthcare institutions;
- the index of utility of some drugs equals "0"; it indicates the absence of the drug in the current State formulary or in unified clinical protocols of medical care for diseases registered in the given healthcare institution.

Therefore, the results of the integrated ABC/VEN-analysis allow to improve the purchasing policy of essential drugs (AV; AE) under the conditions of the total budget deficit, reduce the use of ineffective drugs (AN; BN) and form an effective local formulary.

CONCLUSIONS

The use of ABC/VEN-analysis under the conditions of introduction of the formulary system is extremely important in order to optimize costs for the drug supply of the diagnostic and treatment process and improvement of the efficiency of pharmacotherapy. Its application in the practical work of healthcare institutions can be the real basis for determination of priorities to optimize the supply campaign.

An essential addition of the ABC/VEN-analysis that allows to assess objectively the degree of necessity of using various drugs registered in a healthcare institution for pharmacotherapy of specific diseases is assessment of its utility ($U_V$, $U_E$, $U_N$). The results of the integrated ABC/VEN-analysis with determination of the level of the index of utility of each drug indicate its effectiveness to control the rational use of budgetary funds on the drug supply.

Coclusions according to the results of the integrated ABC/VEN-analysis will allow to optimize the structure of the local formulary of the healthcare institution, provide priority funding for purchasing vital drugs, prepare a substantiated request for the next period of the drug purchase, take appropriate management decisions by the healthcare institution concerning the further implementation of the formulary system principles.

The annual integrated ABC/VEN-analysis conducted by PTC with determination of the level of utility of each drug included into the local formulary allows to identify routinely the problems existing in the healthcare institution in relation to pharmacotherapy of diseases, thereby significantly increasing the level of medical care and fully implementing the medical, pharmacological, economic, professional, educational and informational functions of the formulary system.

REFERENCES


1. *Воробьев П.А. // Проблемы стандартизации в здравоохранении. – 2004. – №7. – С. 82-114.*
2. *Ерофеева С.Б., Соболев К.Э. // Заместитель главного врача. – 2012. – №5. – С. 123-126.*
3. *Звягинцева Т.В., Мирончeнко С.И. // Експеримент. і клін. медицина. – 2011. – №2 (51). – С. 61-66.*
4. *Кабачна А.В., Кабачний О.Г., Шелкова Е.В., Рогова О.Г. Впровадження формулярної системи в практичну роботу лікувально-профілактичних закладів: Метод. рекоменд. – Х.: НФаУ, 2011. – 33 с.*
5. *Кабачна А.В., Шелкова Е.В., Кабачний О.Г. Проведення фармакотерапевтичними комісіями закладів охорони здоров'я аналізу використання бюджетних коштів на лікарське забезпечення: Метод. рекоменд. – Х.: НФаУ, 2014. – 25 с.*
6. *Кабачна А.В., Парфьонова І.І., Шелкова Е.В., Кабачний О.Г. Методика навчання членів фармакотерапевтичних комісій закладів охорони здоров'я принципам проведення ABC/VEN-аналізу раціонального використання бюджетних коштів на лікарське забезпечення за допомогою Microsoft Office Excel: Метод. рекоменд. – Х.: НФаУ, 2014. – 30 с.*
7. *Клинико-экономический анализ / Под ред. П.А.Воробьева. – М.: Ньюдиамед, 2008. – 707 с.*
8. *Морозов А.М., Степаненко А.В., Думенко Т.Ф. Державний формуляр лікарських засобів у системі охорони здоров'я: Монографія. – Чернігів, 2013. – 228 с.*
9. *Попович В.П., Громовик Б.П. // Фармакол. та лікарська токсикол. – 2012. – №2. – С. 59-61.*
10. *Про затвердження шостого випуску Державного формуляра лікарських засобів та забезпечення його доступності: Наказ МОЗ України від 08.04.2014 №252 [Електронний ресурс]. – Режим доступу: http://moz.gov.ua/ua/portal/dn_20140408_0252.html*
11. *Про створення формулярної системи забезпечення лікарськими засобами закладів охорони здоров'я: Наказ МОЗ України від 22.07.2009 №529 [Електронний ресурс]. – Режим доступу: http://moz.gov.ua/ua/portal/dn_20090722_529.html*
12. *Про порядок закупівлі лікарських засобів закладами та установами охорони здоров'я , що фінансуються з бюджету: Постанова Кабінету Міністрів України від 05.09.1996 №1071 (зі змінами) [Електронний ресурс]. – Режим доступу: http://www.moz.gov.ua/ua/portal/dp_19960905_1071.html*
13. *Яковлєва Л.В. // Вісник фармакол. та фармації. – 2007. – №7. – С. 19-23.*
14. *Gupta R., Gupta K., Jain B. // MJAFI. – 2007. – Vol. 63, №4. – P. 325-327. [Електронний ресурс]. – Режим доступу: // http://medind.nic.in/maa/t07/i4/maat07i4p325.pdf*


**ОЦІНКА РАЦІОНАЛЬНОСТІ ВИКОРИСТАННЯ ЗАКЛАДАМИ ОХОРОНИ ЗДОРОВ'Я БЮДЖЕТНИХ КОШТІВ НА ЛІКАРСЬКЕ ЗАБЕЗПЕЧЕННЯ В УМОВАХ ВПРОВАДЖЕННЯ ФОРМУЛЯРНОЇ СИСТЕМИ**

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***Ключові слова*: формулярна система; локальний формуляр; лікарські засоби; ABC/VEN-аналіз; індекс утилітарності; фармакотерапевтична комісія**

*Зростання витрат на лікарське забезпечення в охороні здоров'я вимагає ефективних методів їх аналізу в конкретних закладах охорони здоров'я. На прикладі умовної установи охорони здоров'я проведено інтегрований ABC / VEN-аналіз витрат на лікарське забезпечення за рік з використанням індексу утилітарності кожного лікарського засобу. Результати проведеного аналізу дозволять оптимізувати закупівлю лікарських засобів відповідно до структури захворювань, які реєструються в даному лікувальному закладі, та оптимізувати структуру локального формуляра.*

**ОЦЕНКА РАЦИОНАЛЬНОСТИ ИСПОЛЬЗОВАНИЯ УЧРЕЖДЕНИЯМИ ЗДРАВООХРАНЕНИЯ БЮДЖЕТНЫХ СРЕДСТВ НА ЛЕКАРСТВЕННОЕ ОБЕСПЕЧЕНИЕ В УСЛОВИЯХ ВНЕДРЕНИЯ ФОРМУЛЯРНОЙ СИСТЕМЫ**

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***Ключевые слова:* формулярная система; локальный формуляр; лекарственные средства; ABC/VEN-анализ; индекс утилитарности; фармакотерапевтическая комиссия**

*Рост расходов на лекарственное обеспечение в здравоохранении требует эффективных методов их анализа в конкретных учреждениях здравоохранения. На примере условного учреждения здравоохранения проведен интегрированный ABC/VEN-анализ расходов на лекарственное обеспечение за год с использованием индекса утилитарности каждого лекарственного средства. Результаты проведенного анализа позволяют оптимизировать закупку лекарственных средств в соответствии со структурой заболеваний, которые регистрируются в данном лечебном учреждении, и оптимизировать структуру локального формуляра.*

*Recommended by Doctor of Pharmacy, professor D.I.Dmitriyevsky*

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# PECULIARITIES OF APPLICATION OF THE MODEL OF ADAPTATION OF THE ORGANIZATIONAL STRUCTURE OF PHARMACEUTICAL ENTERPRISES TO THE EXTERNAL ENVIRONMENT CHANGES

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*Key words: model of adaptation; organizational structure; structural subdivisions; factors of the external environment; manufacturing pharmaceutical enterprises*

*Today, a high degree of uncertainty and instability of the external environment determines the growing importance of adaptation of modern organizations to the new realities. For effective functioning of manufacturing pharmaceutical enterprises it is necessary to use the method that allows to describe the processes of transformation of the internal environment in the mode of constant adaptation to changes in the external environment. The peculiarities of application of the model adaptation of the organizational structure of pharmaceutical enterprises to changes in the external environment on the basis of interaction of its subdivisions are presented. The efficiency and speed of the adaptation process of pharmaceutical enterprises are determined by systemic relationship of three matrices: harmonization, which determines the principles and dynamics of interactive planning and evaluation of the results of adaptive transformations; resources determining the rationality of their distribution between the structural subdivisions of the enterprise to implement transformations; structural interaction that determines the optimal interaction based on the in-house staff motivation and other social and psychological factors. The adaptation process has been analyzed for the case when for the change of two essential factors of the external environment the pharmaceutical enterprise reacts by changing its three structural subdivisions. The mechanism of collection and subsequent transfer of information concerning the external environment of the manufacturing pharmaceutical enterprise and its influence on the structural subdivisions is given. It is expedient to use this model for preserving and strengthening the competitiveness of pharmaceutical enterprises at the domestic pharmaceutical market.*

Complications of the market space due to development of competition, information technologies, globalization of business, and a high degree of uncertainty and instability of the external environment determine the growing importance of adaptation of modern organizations to the new realities. For effective functioning of manufacturing pharmaceutical enterprises it is necessary to use the method that allows to describe the processes of transformation of the internal environment in the mode of constant adaptation to changes in the external environment.

In the works of the well-known foreign authors the problems of management of enterprise adaptation to changes of the external environment are considered [5-10]. In scientific professional journals of pharmacy there are articles on modeling the process of adaptation of pharmaceutical enterprises to the impact of the external environment [4]. There is almost no information in literature about the adaptation models of manufacturing pharmaceutical enterprises to changes of the external environment, including model of the organizational structure adaptation to changing external environment components.

The aim of the work is the peculiarities of using the adaptation model of the organizational structure to changes of the external environment on the example of a manufacturing pharmaceutical enterprise.

**Materials and Methods**

The methods of expert assessment, economic and mathematical modeling and graphical method were use in the study.

**Results and Discussion**

To ensure survival, resistant functioning and development of pharmaceutical enterprises in the conditions of changes occurring in the external environment it is necessary to use the model that allows to describe the processes of transformations of the internal environment in the mode of constant adaptation to changes in the external environment.

When studying adaptive features of a company the adaptation is meant the intention of pharmaceutical enterprises to continuously reducing deviation of its position from the requirements dictated by the external environment changes. In this case the inefficient result is both negative (lagging) and positive (advance) deviation of the results of work of the enterprise from a certain value determined by the state of the corresponding environmental factor. The process of adaptive transformations can be represented in the form of economic and mathematical model of the tracking system closed by feedback according to the results of transformations (Fig. 1) [1-4]. The structure of the adaptation model contains the matrix of harmonization (interactive planning),

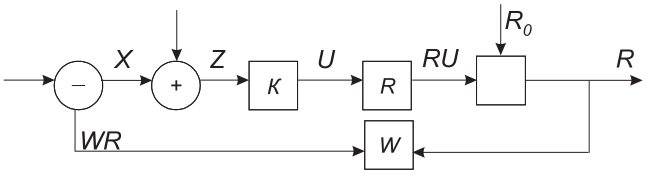


Fig. 1. The structural scheme of the model of adaptation of the organizational structure of pharmaceutical enterprises to changes in the external environment.

the matrix of resources and the matrix of structural interactions.

There are such components in this model:

- *X* − are the most significant environmental factors for pharmaceutical enterprises;
- *K* – is the matrix of resources allocated to perform transformations for each factor of the enterprise internal environment;
- *R* − is the matrix of the structural interaction (the number of variables of the enterprise internal environment subjected to adaptive transformations, namely the number of structural subdivisions of pharmaceutical enterprises);
- W – is the matrix of harmonization (this matrix specifies the structure of the the current state of the enterprise to the requirements of the external environment).

In this model, the complex of information and analytical measures allows management to assess the extent of compliance of the pharmaceutical enterprise state to the requirements dictated by changes in the external environment. It is described by a linear function of coordination $\Delta = X\text{-}WR$ where $\Delta$ – is the value of deviation of the enterprise state from the system of requirements specified by the state of the external environment and determined by some coordination function, which is dependent on measuring the variables of the external and internal environment.

The adaptation process was analyzed for the case when instead of two significant factors of the external environment the pharmaceutical enterprise responds by changing its three structural subdivisions.

Thus, the matrix of harmonization specifies the system of adaptation goals correlating the ratio and the necessary quantitative level of changes. In general, all factors of the internal environment (for example, all structural units or functional blocks) of the enterprise are to be agreed with each factor of the external environment. In the absence, in the opinion of management, of the need to harmonize any internal factor with this factor of the external environment the relevant component of the harmonization matrix is assumed to be zero. The matrix of harmonization shows which variables of the internal environment of the organization (and in what quantitative and qualitative ratio) require changes for an adequate reaction to changes in each factor of the external environment, as well as how the changes of internal variables are interconnected in the system aspect. In this model each line of the matrix resources shows which part of the resources is allocated to the structural element of the enterprise for solving the task of adaptation to each of the factors in the external environment. Thus, the greater number of aspects is involved in transformations, the more active will be the reaction of the organizational system to changes of the external environment. However, apart from the number of the elements involved, the strength of their interactions, the nature of organizational relationships are of great importance, i.e. the content and effectiveness of changes at pharmaceutical enterprises are additionally determined by the type and characteristics of its structure.

In this model, the structural interaction matrix is a table that describes quantitatively the degree of interaction (coordination) of subdivisions of the enterprise in solving problems of adaptation. The degree of interaction of subdivisions can be assessed in arbitrary units ranging from 0 – the complete lack of interaction to 1 – the maximum possible interaction. For our study the information about the degree of interaction of such structural subdivisions of the pharmaceutical enterprise "A" as Production department, Financial department, Marketing department, HR department and Sales department was of interest. The degrees of interactions of subdivisions for the given pharmaceutical enterprise determined by the assessments of the managers of subdivisions are presented in Table.

For the pharmaceutical enterprise "A" the degree of interaction of financial and production structures can be called high (the degree of interaction – 0.8). Finance and marketing functional areas interact the most densely (the degree of interaction – 0.9) and the Financial department with the Sales department (0.9). The internal interaction (self interaction) of each subdivision is maximal (1.0) (the diagonal cells of the matrix). The low interaction is observed between the Production department with the Sales department (0.3) and the HR department (0.3). The degree of interaction of the Marketing and the Production department – 0.3 indicates a low level of implementation of marketing management at the enterprise. However, the lowest is the interaction of the HR department with subdivisions of the enterprise, namely: financial (0.1) and marketing (0.2) services, the Sales department (0.2) and the Production department (0.3).

Depending on the type of the structure and socio-psychological, administrative, technological and other features of the enterprise the structure matrix can take an intermediate form from complete absence of interaction to the ideal interaction. For example, pharmaceutical enterprises that have a functional structure are characterized with a weak interaction of subdivisions in the process of adaptive transformations; a high degree of interaction is typical for matrix and multidimensional structures [1, 7]. For the pharmaceutical enterprise "A" the organizational structure according to the principle of functional departmentalization is typical. A weak interaction of subdivisions leads to manifestation of discrepancies in the goals of the subdivisions with the general purposes of the enterprise. The presence of the interaction leads to excellent results – increase in the ef-

Table

The degrees of interaction of subdivisions at the pharmaceutical enterprise "A"

| | Production department | Financial department | Marketing department | HR department | Sales department |
|---|---|---|---|---|---|
| Production department | 1 | 0.8 | 0.3 | 0.3 | 0.3 |
| Financial department | 0.8 | 1 | 0.9 | 0.1 | 0.9 |
| Marketing department | 0.3 | 0.9 | 1 | 0.2 | 0.8 |
| HR department | 0.3 | 0.1 | 0.2 | 1 | 0.2 |
| Sales department | 0.3 | 0.9 | 0.8 | 0.2 | 1 |

fectiveness of the system at the expense of rational relations and the unidirectionality of actions. This result is manifested in the fact that in the presence of interaction the speed of elimination of inconsistencies by the specific environmental factor will depend on the contribution of each structural subdivision of the enterprise even if no resources are allocated to this department for solving the problem of transformation targeted to this factor.

For effective actuation of the model of adaptation of pharmaceutical enterprises to changes in the external environment on the basis of interaction of its subdivisions it is necessary to create the appropriate organizational and economic preconditions. To do this at first the organizational form for conducting the analysis of the external environment should be specified. When studying specific areas of the external environment the company may apply to specialized research organizations, such as consulting and information firms, scientific research institutes, higher schools, and conduct research on their own. For the pharmaceutical enterprise "A" the introduction of the model of adaptation of the organizational structure to changes in the external environment is imposed on employees of the Marketing department, they have significant internal resources to perform this task.

The first stage of the model is to monitor environmental factors taking into account the possible information uncertainty. When performing this kind of work it is advisable to combine efforts of the Marketing department with other departments involved in the study of certain aspects of the external environment (Fig. 2).

Therefore, it is necessary to divide powers and responsibilities between the subdivisions in the system of studying the enterprise external environment, as well as to establish information interaction in such a way that there is no duplication of works and at the same time without the information gaps in any areas. This primarily concerns the interaction of the Sales department with the Financial department and the Production department. And the Marketing department should not only collect information from the subdivisions, but also coordinate and direct their efforts to more active data collection concerning the external environment. It is necessary to develop and give recommendations to employees of the enterprise about what, where and from who the information should be collected. The activity in researching the external environment should be carried out with the active support of the top management, which must provide the Marketing department with the open access to information resources of the company, including confidential ones, as well as provide it with the information obtained from its own channels.

All the information collected, processed and analyzed should be stored in databases of customers, suppliers, competitors, contact audiences and macroenvironmental factors. Databases should be designed in a way that could provide information about the environment to different users with the necessary level of detail to them. All databases should be interrelated and must be updated as new information becomes available, particularly in the monitoring process. The final stage of the analysis of the external environment is preparation and presentation of the report to top management of the pharmaceutical enterprise and its subdivisions.

The next step is to analyze the information, and it is the responsibility of the Marketing department. The results of the analysis of deviation of the pharmaceutical enterprise state from the requirements dictated by changes in the external environment are provided by the Marketing department to the heads of the Production department, Financial department, Sales department and other subdivisions and management (CEO, commercial director, director of quality, director of production and CFO). After that the management decision to implement adequate transformations in the internal environment of pharmaceutical enterprises is made by top management and the heads of the departments involved in adaptive changes of the internal environment in a constant adaptation to changes in the external environment. To adapt pharmaceutical enterprises to changes in the external environment it is necessary to allocate the appropriate resources (information, financial, etc.). In this case, all costs for adaptive transformations of pharmaceutical companies should be viewed as investments in pharmaceutical enterprises that promote timely elimination of dangers and use of opportunities, which benefits can have a long-lasting character.

For example, the pharmaceutical enterprise "A" solves the problem of creating a new area for drug manufacturing certified in accordance with the requirements of GMP in order to adapt to consumer new demands for quality and cost of the products. In the process of transformations the production effectively masters allocated material, financial and information resources (Sales department, Marketing department and Financial

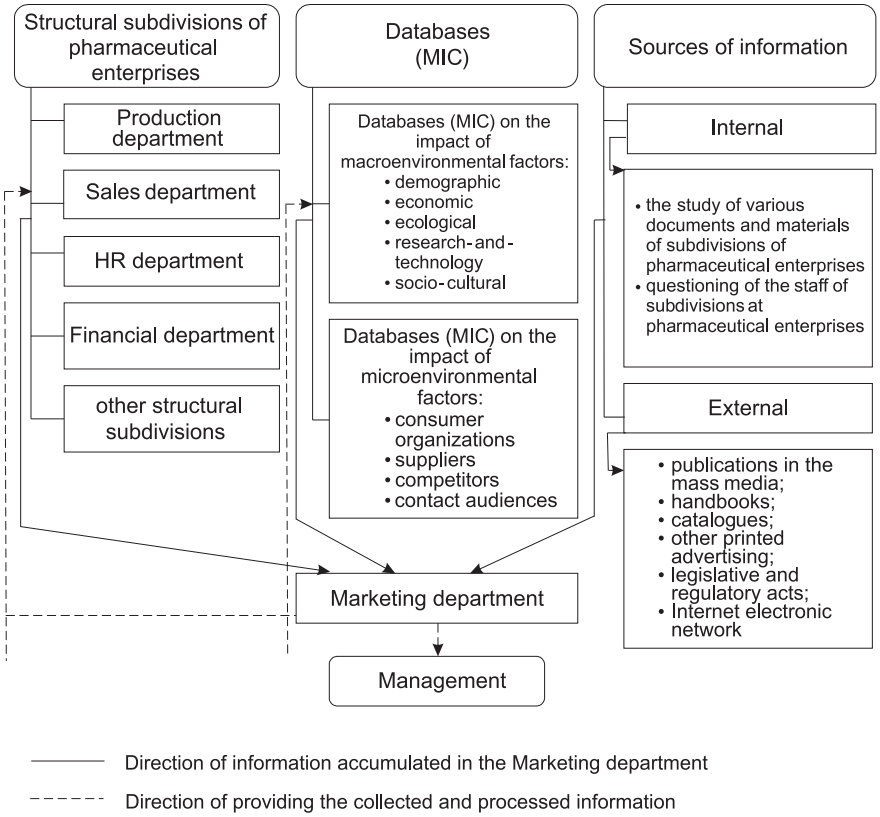


Fig. 2. The scheme of collection and subsequent transfer of information concerning the external environment of the pharmaceutical enterprise "A" and its influence on the structural subdivisions.

department). As part of this transformation the HR department of the enterprise is tasked regarding recruitment of the staff with the skills needed for work on the new equipment. The presence of effective communications in the form of horizontal and diagonal (including informal) relationships between production and the HR department allows managers of these subdivisions to coordinate additionally the requirements to potential employees reasonably going beyond the formal job descriptions. Under these conditions the work of HR will be more focused because it would better consider the requirements of the "customer", and it improves the quality of selection and reduces the time to find candidates by the head of the Production department. Thus, the interaction between two subdivisions provides multiplication of efforts and the unidirectionality of actions increasing the response rate of the entire enterprise.

The head of the Marketing Department will be responsible for the control and operation of the model developed. The heads of departments together with employees of the Marketing department will provide the legal support for the model.

It is expedient to use this model for preserving and strengthening the competitiveness of pharmaceutical enterprises at the domestic pharmaceutical market.

CONCLUSIONS

1. The peculiarities of application of the model of adaptation of the organizational structure of pharmaceutical enterprises to changes in the external environment, which appropriately describes the process of transformation of the internal environment of the organization in a constant adaptation to environmental changes, are given.

2. The efficiency and speed of the adaptation process of pharmaceutical enterprises are determined by systemic relationship of three matrices: harmonization, which determines the principles and dynamics of interactive planning and evaluation of the results of adaptive transformations; resources determining the rationality of their distribution between the structural subdivisions of the enterprise to implement transformations; structural interaction that determines the optimal interaction based on the in-house staff motivation and other social and psychological factors.

REFERENCES

1. *Бондарєва І.В. Актуальні питання створення нових лікарських засобів // Матер. Всеукр. наук.-практ. конф. студ. та молодих учених, 23-24 квітня 2009 р., м. Харків. – Х.: Вид-во НФаУ, 2009. – С. 308.*
2. *Бондарєва І.В. Актуальні питання створення нових лікарських засобів // Матер. Всеукр. наук.-практ. конф. студ. та молодих учених, 21-22 квітня 2010 р., м. Харків. – Х.: Вид-во НФаУ, 2010. – С. 345.*
3. *Михненко П.А. // Менеджмент в России и за рубежом. – 2008. – №3. – С. 3-10.*

4. *Пестун І.В. Моделювання процесу адаптації фармацевтичного підприємства до змін зовнішнього середовища: Метод. рекоменд. / І.В.Пестун, І.В.Бондарєва. – Х.: Вид-во НФаУ, 2010. – 24 с.*
5. *Frishammar J. // Management Decision. – 2003. – Vol. 41, №4. – P. 318-326.*
6. *Hough J. // Management Decision. – 2004. – Vol. 42, №6. – P. 781-793.*
7. *Karim N. // Library Review. – 2004. – Vol. 53, №7. – P. 356-362.*
8. *Velitchka D. // J. of Marketing. – 2006. – Vol. 70, №1. – P. 107-118.*
9. *Walsh P. // Management Decision. – 2005. – Vol. 43, №1. – P. 113-122.*
10. *Yunggar M. // J. of American Academy of Business, Cambridge. – 2005. – Vol. 6, №2. – P. 324-331.*

**ОСОБЛИВОСТІ ЗАСТОСУВАННЯ МОДЕЛІ АДАПТАЦІЇ ОРГАНІЗАЦІЙНОЇ СТРУКТУРИ ФАРМАЦЕВТИЧНИХ ПІДПРИЄМСТВ ДО ЗМІН ЗОВНІШНЬОГО СЕРЕДОВИЩА**

***І.В.Бондарєва***

***Ключові слова:*** *модель адаптації; організаційна структура; структурні підрозділи; фактори зовнішнього середовища; виробничі фармацевтичні підприємства*

*На теперішній час високий ступінь невизначеності та нестабільності зовнішнього середовища обумовлює зростання значущості адаптації сучасних організацій до нових реалій. Для ефективного функціонування виробничих фармацевтичних підприємств необхідно використання методики, що дозволяє в режимі постійної адаптації до змін зовнішнього середовища описати процеси перетворень внутрішнього середовища. Надані особливості застосування моделі адаптації фармацевтичних підприємств до змін зовнішнього середовища на засадах взаємодії його підрозділів. Ефективність і швидкість процесу адаптації фармацевтичних підприємств визначаються системним взаємозв'язком трьох матриць: узгодження, яка визначає принципи і динаміку інтерактивного планування і оцінювання результатів адаптаційних перетворень; ресурсів, яка визначає раціональність їх розподілу між структурними одиницями підприємства для здійснення перетворень; структурної взаємодії, що визначає оптимальну внутрішньоорганізаційну взаємодію з урахуванням мотивації співробітників та інших соціально-психологічних чинників. Проаналізовано процес адаптації для випадку, коли на зміну двох суттєвих факторів зовнішнього середовища фармацевтичне підприємство реагує зміною трьох його структурних підрозділів. Надано механізм збору і подальшої передачі інформації стосовно зовнішнього середовища на виробничому фармацевтичному підприємстві та його впливу на структурні підрозділи організації. Використання даної моделі є доцільним для збереження і посилення конкурентоспроможності фармацевтичних підприємств на вітчизняному фармацевтичному ринку.*

**ОСОБЕННОСТИ ПРИМЕНЕНИЯ МОДЕЛИ АДАПТАЦИИ ОРГАНИЗАЦИОННОЙ СТРУКТУРЫ ФАРМАЦЕВТИЧЕСКИХ ПРЕДПРИЯТИЙ К ИЗМЕНЕНИЯМ ВНЕШНЕЙ СРЕДЫ**

***И.В.Бондарева***

***Ключевые слова:*** *модель адаптации; организационная структура; структурные подразделения; факторы внешней среды; производственные фармацевтические предприятия*

*Сегодня высокая степень неопределенности и нестабильности внешней среды обусловливает рост значимости адаптации современных организаций к новым реалиям. Для эффективного функционирования производственных фармацевтических предприятий необходимо использование методики, позволяющей в режиме постоянной адаптации к изменениям внешней среды описывать процессы преобразований внутренней среды. Представлены особенности применения модели адаптации фармацевтических предприятий к изменениям внешней среды на основе взаимодействия его подразделений. Эффективность и скорость процесса адаптации фармацевтических предприятий определяются системной взаимосвязью трех матриц: согласования, которая определяет принципы и динамику интерактивного планирования и оценки результатов адаптационных преобразований; ресурсов, которая определяет рациональность их распределения между структурными единицами предприятия для осуществления преобразований; структурного взаимодействия, которая определяет оптимальное внутриорганизационное взаимодействие с учетом мотивации сотрудников и других социально-психологических факторов. Проанализирован процесс адаптации в случае, когда на смену двух существенных факторов внешней среды фармацевтическое предприятие реагирует изменением трех его структурных подразделений. Предоставлен механизм сбора и последующей передачи информации о внешней среде на производственном фармацевтическом предприятии и ее влияния на его структурные подразделения. Использование данной модели целесообразно для сохранения и усиления конкурентоспособности фармацевтических предприятий на отечественном фармацевтическом рынке.*

*Recommended by Doctor of Pharmacy, professor A.A.Kotvitska*

UDC 615.15: 614.27

# THE STUDY OF MODERN APPROACHES TO PROVISION OF PHARMACEUTICAL CARE IN THE CONDITIONS OF GOOD PHARMACY PRACTICE INTRODUCTION

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*Key words: pharmaceutical care; pharmaceutical service; pharmacy practice; pharmacotherapy; system (comprehensive) approach*

*It has been shown that nowadays pharmaceutical provision of the population is a complex integration system that combines pharmaceutical care, pharmaceutical services, the process of drug supply and professional pharmacy practice. The content of such concepts as pharmaceutical care and pharmaceutical service, as well as medication therapy management have been determined. Two approaches to the management of pharmaceutical services in the process of changing the vector of the professional activity of a pharmacist have been reviewed. With regard to the implementation of such services there are two approaches: a focus on a product (drug) and focus on a patient. The basic functional differences between these two approaches in providing pharmaceutical services have been identified. A comparative analysis of the parameters for such concepts as pharmaceutical and medical care has been carried out. It has been shown that the process of medication therapy management includes three levels: system (public policy, regulation and legislation), institutional (control lists, formularies and protocols) and individual (pharmaceutical care). From positions of the system approach the process of pharmaceutical care by the patient's treatment planning has been considered.*

In 1998 FIP was first adopted the "Regulations on professional standards of pharmaceutical care". It offers guidance for national healthcare systems (NHS) regarding implementation of pharmaceutical care. The new paradigm of good pharmacy practice (GPP) is presented in the joint publication of the WHO and FIP "Developing pharmacy practice. A focus on patient care". In 2011 FIP jointly with the WHO adopted an updated version of good pharmacy practice "Joint FIP/WHO guide to good pharmacy practice: standards of pharmacy services quality" [1, 2, 4].

There is a tendency in the world to shift the focus of a pharmacist initially focused only on medicines in providing of pharmaceutical care (PC) to a patient. Therefore, the role of the pharmacist has evolved, and he has turned from someone who prepares and sells medicines on the provider of service and information [1, 5]. The complexity of the evolutionary processes of changing the role of pharmacists in the pharmacy practice makes it particularly relevant to study the approaches for providing PC to patients [1, 7, 9].

**Materials and Methods**

The analysis of domestic and foreign sources has revealed that at present there are no analytical system studies of peculiarities for providing PC to the patients in pharmacies [2-6, 10]. In this context, the aim of the study was a systematic analysis of approaches of providing PC to patients in the pharmacy practice. The objects of the study were scientific concepts and terms directly related to the PC provision and professional practice.

**Results and Discussion**

Pharmaceutical service involves all kinds of services provided by pharmacists in the framework of PC. Together with dispensing drugs such services also include informing, education and counseling. Pharmaceutical service is PC given to the population as a result of the pharmacist professional practice that have cost estimate (based on contractual prices).

Analysis of the modern approaches to the PC provision needs to consider the concept of pharmaceutical service associated with the English term "medication therapy management". This term was introduced in the USA in connection with the activities of the federal health insurance programme Medicare for seniors (2006). The term "medication therapy management" is defined as professional activities, in which the help is given to a patient according to the certain standards that ensure each patient to use drugs individually corresponding to the medical case and have an effective and safe therapy using the system (comprehensive) approach [5].

The term "system" is characteristic of the American approach and means the provision of medical and pharmaceutical care to patients in order to achieve the necessary results of individual therapy and coordinate medical and pharmaceutical professionals. This change of the professional activities vector is related with the existence of two approaches for the so-called pharmaceutical service management. From the point of view of implementation of such services, there are two approaches: focus on a product (drug) and focus on a patient [4, 10, 11]. The main differences of both approaches are shown in Fig. 1.

The first of these approaches is traditional for many countries and is associated with the process of drug

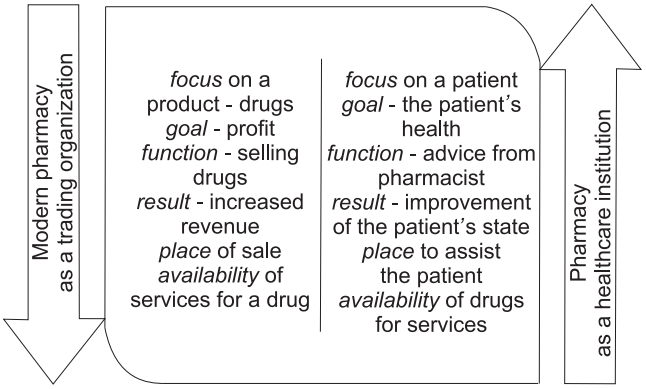


Fig. 1. The main functional differences between two approaches in providing pharmaceutical services.

distribution from the manufacturer to the patient, i.e. the pharmacy performs its commercial function in order to obtain the maximum profit. The focus is clearly aimed at selling the product with occurrence of many economic and ethical conflicts of interests that lead to many negative effects, such as polypragmasy. Usually a patient in this approach is not at the first place, and the problem of appropriate drug therapy and pharmacy practice is inferior to business interests.

The second approach separates the process of providing pharmaceutical services from the process of drug sales and determines it as a resource to achieve the ultimate goal. As part of this approach the activity of the professionally trained specialist such as a clinical pharmacist in hospitals or outpatient services or a pharmacist in an ordinary pharmacy giving consultations is considered. Pharmaceutical services are based on professional PC practice, which provides individual level of service, and is regarded as one of the types of care within the concept of a self-care patient. A pharmacist within PC is responsible for decisions concerning the results of therapy that should be effective and safe. This leads to the conclusion that such a service is an independent process and does not replace the role of doctors.

Medication therapy management involves three levels: system (public policy, regulation and legislation), institutional (regulatory lists, formularies and reports) and individual level. PC can be directed both to individuals and to groups of the population.

Within the "PD-oriented to the population" in order to create formularies or lists of drugs one can use demographic and epidemiological data, develop the regulatory framework for the pharmaceutical sector and monitor its practical introduction, create and manage pharmacy networks, prepare and analyze reports as for drug administration, evaluate the use of drugs and inform health professionals about standards and algorithms for the activities of pharmaceutical institutions.

Without individual PC no healthcare system is able to provide effective drug therapy and the monitoring of its results arising from the use of drugs. These measures implemented at the population level are carried out before or after the medical examination of the patient and obtaining relevant information from him, but they can not replace pharmaceutical services focused on a patient and provided with direct contact. Patients need the services of a pharmacist at the time of medical care.

Successful pharmacotherapy is individual for each patient. It involves individual approach in decision-making concerning the drug therapy model choice, achieving concurrence (agreed position between a patient and a professional providing care regarding therapeutic outcomes and ways to achieve them), as well as very important measures on monitoring of the patient's condition.

The system approach to providing individual PC to a patient is shown in Fig. 2. For individual treatment of each individual patient a pharmacist develops a treatment plan with him/her.

Therefore, patients can make their contribution to the success of therapeutic results taking a share of responsibility for their own treatment and not resting it entirely on healthcare professionals as it was done previously within the paternal approach.

The results of comparison of the parameters of the concepts of medical and pharmaceutical care are given in Table.

In this context, the role of the pharmacist in the healthcare system is fundamentally changing. Practice of PC is new and significantly different from what was the activity of pharmacists in recent years. Thus, the cor-

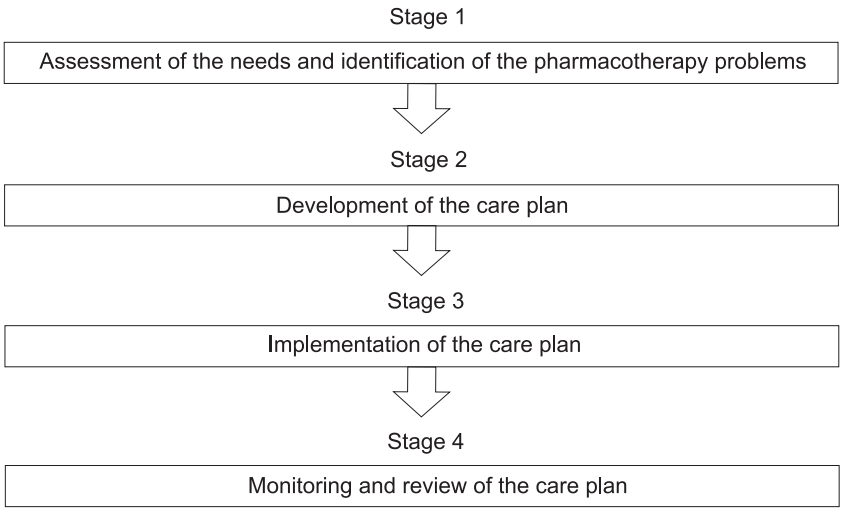


Fig. 2. The system approach to providing pharmaceutical care to a patient.

Table

A comparative analysis of parameters of medical and pharmaceutical care

| The type of care | Primary focus | Basic knowledge | A decision on using a drug |
|---|---|---|---|
| Medical | Diagnosis and treatment of a patient | Pathophysiology | Prescribing the drug |
| Pharmaceutical | Identification and satisfaction of the patient's needs in medications | Pharmacotherapy | Prevention and solution of problems associated with pharmacotherapy |

nerstone in modern practice of PC providing is a professional responsibility of a pharmacist.

CONCLUSIONS

The study of approaches for providing PC and their scientific generalization are important and relevant for development of good pharmacy practice in Ukraine. Modern PC should be considered as pharmacy practice that involves the professional responsibility of a pharmacist for implementation of pharmaceutical services with the focus on a patient.

REFERENCES


1. *Дремова Н.Б., Коржавых Э.А., Литвинова Т.М. и др. // Фармация. – 2005. – №2. – С. 37-45.*
2. *Належна аптечна практика. Стандарти якості аптечних послуг: Спільна настанова МФФ/ВООЗ від 01.01.2011 р. Режим доступу до настанови: http://zakon2.rada.gov.ua/laws/show/897_009.*
3. *Панфілова Г.Л., Немченко А.С., Немченко О.А. Організація фармацевтичної допомоги населенню в умовах медичного страхування. – Х.: Ависта-ВЛТ, 2009. – 226 с.*
4. *Розвиток фармацевтичної практики: фокус на пацієнта. Настанова ВООЗ, 2006 (Developing pharmacy practice – A focus on patient care. WHO Guideline, 2006). – 65 с.*
5. *Bluml B. // J. Am. Pharm. Assoc. – 2005. – Vol. 45. – P. 566-572.*
6. *Cousins D., Kijlstra N., Walser S. Pharmaceutical Care: Policies and Practices for a Safer, More Responsible and Cost-effective Health System. – Strasbourg: EDQM, 2012. – 62 p.*
7. *Hepler C.D., Strand L.M. // Am. J. Hosp. Pharm. – 1990. – Vol. 47. – P. 533-43.*
8. *Medication therapy management in pharmacy practice: core elements of an MTM service model (version 2.0) // J. Am. Pharm. Assoc. – 2008. – Vol. 48. – P. 341-353.*
9. *Rickles N.M., Wertheimer A.I., Smith M.C. Social and Behavioral Aspects of Pharmaceutical Care. 2-nd ed. – Boston: Jones & Bartlett Publishers, 2010. – 484 p.*
10. *Rovers J.P., Currie J.D. A Practical Guide to Pharmaceutical Care. – Washington: APhA, 2012. – Режим доступу: http://www.pharmacist.com/learning-aids-practical-guide-pharmaceutical-care-3rd-edition.*
11. *Strand L., Cipolle R., Cipolle R.J. et al. Pharmaceutical Care Practice: The Patient-Centered Approach to Medication Management. 3-rd ed. – New York: McGraw Hill Medical, 2012. – 704 p.*


**ДОСЛІДЖЕННЯ СУЧАСНИХ ПІДХОДІВ ДО НАДАННЯ ФАРМАЦЕВТИЧНОЇ ДОПОМОГИ В УМОВАХ ВПРОВАДЖЕННЯ НАЛЕЖНОЇ АПТЕЧНОЇ ПРАКТИКИ**

***В.М.Назаркіна, О.А.Немченко***

***Ключові слова:*** *фармацевтична допомога; фармацевтична послуга; аптечна практика; фармакотерапія; системний підхід*

*Показано, що на теперішній час фармацевтичне забезпечення населення є складною інтеграційною системою, яка поєднує фармацевтичну допомогу, фармацевтичні послуги, процес забезпечення ліками та професійну аптечну практику. Визначено зміст понять фармацевтичне обслуговування та фармацевтична послуга, а також medication therapy management. Розглянуті два підходи до менеджменту фармацевтичних послуг у процесі зміни вектора професійної діяльності фармацевта. З точки зору здійснення таких послуг виділяються два підходи: фокус на товар (Drug) та фокус на пацієнта. Встановлені основні функціональні відмінності обох підходів в наданні фармацевтичних послуг. Виявлені переваги сучасних фармацевтичних послуг, орієнтовані на пацієнта: фармацевт несе відповідальність за результати фармакотерапії, яка має бути ефективною та безпечною. Здійснено порівняльний аналіз параметрів понять фармацевтична та медична допомога. Показано, що процес Medication therapy management передбачає три рівні: системний (державна політика, регулювання та законодавство), інституційний (регулюючі переліки, формуляри та протоколи) та індивідуальний (фармацевтична допомога). З позицій системного підходу розглянуто процес надання фармацевтичної допомоги шляхом складання плану лікування пацієнта.*

**ИССЛЕДОВАНИЕ СОВРЕМЕННЫХ ПОДХОДОВ К ОКАЗАНИЮ ФАРМАЦЕВТИЧЕСКОЙ ПОМОЩИ В УСЛОВИЯХ ВНЕДРЕНИЯ НАДЛЕЖАЩЕЙ АПТЕЧНОЙ ПРАКТИКИ**

***В.Н.Назаркина, О.А.Немченко***

***Ключевые слова:*** *фармацевтическая помощь; фармацевтическая услуга; аптечная практика; фармакотерапия; системный подход*

*Показано, что в настоящее время фармацевтическое обеспечение населения является сложной интеграционной системой, которая сочетает фармацевтическую помощь, фармацевтические услуги, процесс обеспечения лекарствами и профессиональную аптечную практику. Определено содержание понятий фармацевтическое обслуживание и фармацевтическая услуга, а также medication therapy management. Рассмотрены два подхода к менеджменту фармацевтических услуг в процессе изменения вектора профессиональной деятельности фармацевта. С точки зрения осуществления таких услуг выделяются два подхода: фокус на товар (Drug) и фокус на пациента. Установлены основные функциональные различия обоих подходов в предоставлении фармацевтических услуг. Выявлены преимущества современных фармацевтических услуг, ориентированных на пациента: фармацевт несет ответственность за результаты фармакотерапии, которая должна быть эффективной и безопасной. Осуществлен сравнительный анализ параметров понятий фармацевтическая и медицинская помощь. Показано, что процесс Medication therapy management предусматривает три уровня: системный (государственная политика, регулирование и законодательство), институциональный (регулирующие перечни, формуляры и протоколы) и индивидуальный (фармацевтическая помощь). С позиций системного подхода рассмотрен процесс предоставления фармацевтической помощи путем составления плана лечения пациента.*

*Recommended by Doctor of Pharmacy, professor A.A.Kotvitska*

UDC 338.45

# DEVELOPMENT OF METHODOLOGICAL FRAMEWORK FOR SETTING THE DRUG STOCK IN PHARMACY DISTRIBUTION

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*Key words: personalization; logistic service; customer; social responsibility*

*The article has proven that the use of the traditional approach to setting the drug stock level in pharmacy distribution can lead to determination of the level of money immobilized for stock making and does not take into account the needs, expectations and peculiarities of customers, as well as different social significance of specific pharmacological groups of drugs. The methodology making basis for the existing approach to the stock setting does not meet the current requirements to the proper level of the customer service. Thus, the need to improve the approach to setting the medication stock level in pharmacy distribution has arisen. The social responsibility of pharmaceutical business foresees the use of both economic and social parameters in drug stock setting. In social terms, the stock setting task is to determine a list and volume of medications to be available at a warehouse to meet the customers' needs. The stock should have the following groups: quarantine-prevention, current, buffer and reserve. The volume of daily sale of medications is calculated taking into account the volume of sale: in case of the periods with no seasonal demand fluctuations taking into account the trend of changes in frequency of prescribing drugs in nosology; the reserve stock of medications in case of the unforeseen demand fluctuations; in the period of seasonal demand fluctuations. It has been proven that it is reasonable to substantiate the stock level in pharmacy distribution using the ABC-VEN analysis.Meeting the customer's needs is the main task of any pharmacy company, including wholesalers. So, it needs to make a stock of pharmaceuticals neither too small – since the company would not be able to maintain the sales and respond flexibly to demand fluctuations, nor too large – it may cause the increase in storage costs, turnover decrease, 'freezing' of money invested into purchase. Setting of the stock rate is used to determine the necessary stock level.*

The necessity of stock setting to improve the stock management is explained in the works of Alesynska T., Barkalov S., Hadzhynskyi O., Kalchenko A., Kindii M., Krykavskyi Ye., Mate T., Novikov O., Oklander M., etc. The analysis of their research has shown that the stock setting methodology is a product of a daily stock consumption by the stock level. The same approach is traditionally used when setting the stock of medications in pharmacy distribution. These issues are dealt with in the works of Hromovyk B., Gudzenko O., Mnushko Z., Nemchenko A., Tolochko V., Trokhymchuk V., Barnatovych S., Dorokhova L., Gorbunova O., Kutsenko S. and others.

**Experimental Part**

Traditional approaches to setting the drug stock level in pharmacy distribution have a general nature being more characteristic of the level of money immobilized for stock making, and without taking into account the needs, expectations and peculiarities of customers. The methodology making basis for the existing approach to the stock setting does not meet the current requirements to the proper level of the customer service. Thus, the need to improve the approach to setting the medication stock level in pharmacy distribution has arisen.

The aim of this article is to develop the methodological framework for setting the stock of medications in pharmacy distribution based on the differentiation of the customer logistic service.

**Materials and Methods**

The comparative analysis methods, statistical and analytical method of stock setting were used in this research.

**Results and Discussion**

The social responsibility of pharmaceutical business foresees the use of both economic and social parameters in drug stock setting.

The cost-efficiency principle suggests reaching of the rational (optimal) cost-income ratio in the company's performance. The wholesale pharmaceutical companies (WPC) implement this principle by choosing the most profitable medications and health products for sale or the products that have the highest cost contribution rate.

The social responsibility principle means the company's commitment to its product or service. The intense competition at the pharmaceutical distribution market requires distributors to develop individual logistic solutions to meet the customers' needs. Therefore, the WPC must have socially important medications regardless of their profitability.

In economic (financial) terms, the stock setting task is to determine an amount of money that may be immobilized from the company's turnover. In social terms, the stock setting task is to determine a list and volume of medications to be available at a warehouse to meet the customers' needs.

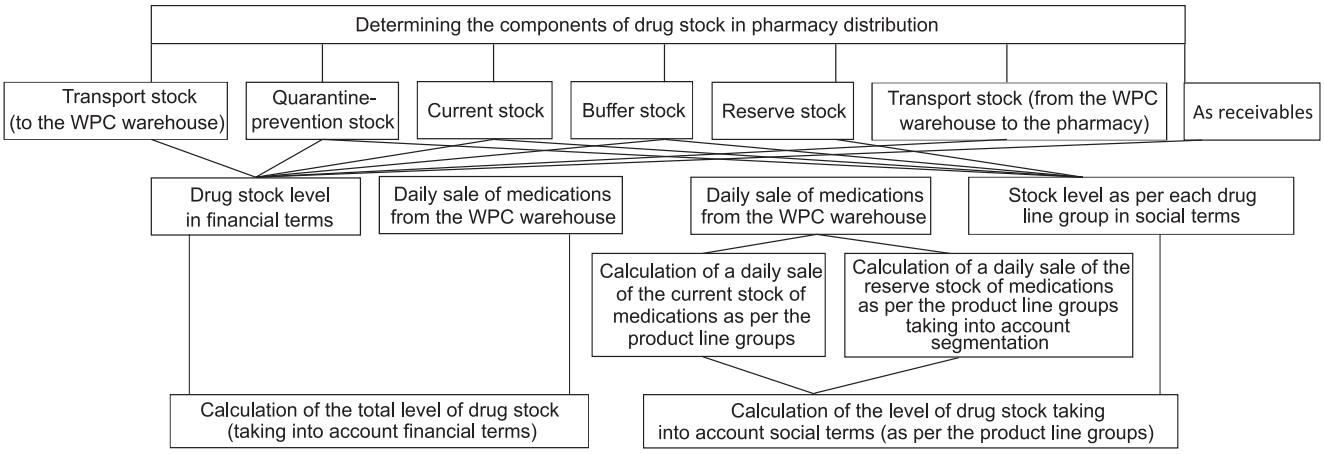


Fig. The scheme proposed for calculation of drug stock in pharmacy distribution taking into account the financial and social aspects.

The level with these two parameters (economic and social) is calculated with the known formula: H = O × D where O is a daily sale of stock, D is a stock level. But the approaches to determine the daily sale of drug stock from the WPC warehouse and the stock level must be different (Fig.).

Different aspects of stock setting determine a set of groups of drug stock in the pharmacy distribution. In social terms, the stock should have the following groups: quarantine-prevention ( $T^s_{k\text{-}p}$); current ( $T^s_c$); buffer ( $T^s_b$); reserve ( $T^s_r$). Thus, taking into account the social parameters the total stock level will be:

$$T^s = T^s_{q\text{-}p} + T^s_c + T^s_b + T^s_r . \quad (1)$$

Such limits and grouping of stock are explained by the peculiarities of processes and activities performed by the WPC to meet the customers' needs. In financial terms, the stock has the following groups: transport (to the WPC warehouse) ( $T^f_{tr}$), quarantine-prevention ( $T^f_{k\text{-}p}$); current ( $T^f_c$); buffer ( $T^f_b$); transport (from the WPC warehouse to the pharmacy) ( $T^f_t$); as receivables ( $T^f_r$). Thus, taking into account the financial parameters the total stock level at the WPC will be:

$$T^f = T^f_{tr} + T^f_{k\text{-}p} + T^f_c + T^f_b + T^f_t + T^f_r . \quad (2)$$

A specific feature of the stock level calculation taking into account the social aspect is a personal approach to the customer servicing that requires to consider the drug status per each product line group. This approach affects both the stock level and calculation of a daily sale.

The social component of the quarantine-prevention stock is made in order to prepare medications for the customer's order kitting. According to the requirements of Good Storage Practice (GSP) [2], all medications at the WPC warehouse must be checked. If a medication does not comply with set quality requirements, such medication must be destroyed or returned to a supplier. If a sale permit is obtained, the medication becomes a stock, is recorded in balance account 'Goods' and transferred to be prepared for the storage area. The preparation stock standard includes the following activities: drug stock transfer from the quarantine area to the acceptance area, drug recording, transfer to the storage area, placing in the storage area. This standard is determined by timing observation and is the same for all drug line groups.

The social component of the current stock of medications at the WPC warehouse is made to ensure the uninterrupted meeting of customer's demand and is calculated as per the product line group. In case of the reliable supply (the rate of the supplier's reliability approaches 1), the current stock is calculated according to the formula:

$$T^s_c = \frac{D}{K_{j\,\mathrm{del}}}, \quad (3)$$

where: D is the number of days in the previous period (month); $K_{jdel}$ is the number of deliveries of the j-drug line group during the period.

The social component of the buffer stock is made to meet the customer's demand in case of an interruption of drug supply to the WPC warehouse. Its level must be calculated taking into account the supplier's reliability rate according to the formula:

$$T^s_b j = \frac{\sum V_{ji} \times (1 - H^n_i) \times T^n_i}{V_j}, \quad (4)$$

where: $V_{ji}$ is delivery of the j-drug line group by the i-th supplier, UAH; $V_j$ is delivery of the j-drug line group by all suppliers, UAH; $T^n_i$ is the delivery time of the j-drug line group by the i-th supplier in case of the reliable supply.

$H^n_i$ is the supplier's reliability rate of the i-th supplier calculated according to the formula [3]:

$$H_n = (1 - P^1_i) \times (1 - P^2_i) \times (1 - P^3_i) \times (1 - P^4_i), \quad (5)$$

where: $p^1_i$ is probability of failure to meet transport terms by the i-th supplier; $p^2_i$ is probability of failure to meet the delivery time; $p^3_i$ is probability of failure to meet the requirements of accompanying documents execution; $p^4_i$ is probability of noncompliance of accompanying documents information with the actual state.

Table 1

Pharmacy Location Segmentation

| Group | Characteristics of a pharmacy | The share of relatively expensive medications, % | The share of relatively cheap medications, % |
|---|---|---|---|
| A | Popular (in the populated areas) | 25-35 | <10 |
| B | Mixed | ≤25 | 10-20 |
| C | Bedroom (in a bedroom district) | ≤20 | 25 |

The social component of the reserve stock is made at WPC warehouses for unforeseen demand fluctuations ( $T^s_{uf}$) and seasonal demand fluctuations ( $T^s_{sf}$).

It is appropriate to calculate the social component of the reserve stock according to the formula:

$$T^s_r = T^s_{uf} + T^s_{sf}. \quad (6)$$

The WPC must always have the drug reserve stock for unforeseen demand fluctuations at the warehouse, it should be sufficient for a period between two subsequent deliveries, it is calculated according to the formula (3). The drug reserve stock for seasonal demand fluctuations should be calculated based on the estimated trends of the seasonal increase in sale of this or that medication in previous years.

In social terms of stock management, the level of a daily sale of medications from the WPC warehouse should be made as per the product line group based on the pharmacy location segmentation. The pharmacy location segmentation methodology was proposed by Slavych-Prystupa O.S. [4] (Tab. 1).

The daily sale of medications from the WPC warehouse taking into account the social component in periods with no seasonal demand fluctuations is calculated as follows:

$$O^s_{j3} = \sum_{n=1}^{3} (O^s_{j\,cn} + O^s_{j\,uf\,n}), \quad (7)$$

where n is a segment of pharmacies; group A is 1; group B is 2; group C is 3: *j* is the number of drug line groups; $O^s_{jcn}$ is a daily sale of the current stock of the j-drug line group for pharmacies of the n-th segment, UAH, calculated according to the formula:

$$O^s_{j\,cn} = \sum_{j=1}^{N} k_j \times \frac{O^{d-1}_{jn}}{D}, \quad (8)$$

where: $k_j$ is a change in drug sale in the n-th segment of pharmacies; it is calculated based on the trend of changes in frequency of prescribing drugs in nosology; $O^{d-1}_{jn}$ is the volume of sales of the *j*-drug line group in the n-th segment of pharmacies in the previous period (month), thousands UAH; *D* is the number of days in the previous period; $O^s_{j\,uf\,n}$ is a daily sale of the reserve stock of the j-drug line group in the n-th segment of pharmacies in case of unforeseen demand fluctuation, UAH.

It has been suggested to use the ABC-VEN analysis to determine the stock for unforeseen demand fluctuation. The matrix of drug grouping according to this method is described in Tab. 2.

Table 2

The matrix of drug grouping according to the ABC-VEN analysis

| Category of stock | V | E | N |
|---|---|---|---|
| A | AV | AE | AN |
| B | BV | BE | BN |
| C | CV | CE | CN |

Table 3

The volume of the drug reserve stock suggested for unforeseen demand fluctuation

| Group | Requirement condition | Reserve stock requirement, % |
|---|---|---|
| AV, AE, BV, CV | P=1 | 100 |
| AN, BE, CE | P=0.95 | 95 |
| BN, CN | Based on the stock shortage analysis | 10-15 |

The requirements to the volume of drug reserve stock at the WPC for unforeseen demand fluctuation as suggested based on the experts survey are given in Tab. 3.

In case of seasonal fluctuations in drug sale the daily volume of sale from the warehouse is calculated according to the formula:

$$O^s_{j3} = \sum_{n=1}^{3} (O^s_{j\,cn} + O^s_{j\,uf\,n} + O^s_{j\,sf\,n}), \quad (9)$$

where: $O^s_{j\,sf\,n}$ is a daily sale of the j-drug line group for pharmacies of the n-th segment; it is calculated according to the formula:

$$O^s_{j\,sf\,n} = \frac{V_{j\,sf\,n}}{T} \times I, \quad (10)$$

where: $V_{j\,sf\,n}$ is the volume of sale of the j-drug line group for pharmacies of the n-th segment during the seasonal demand fluctuation, UAH; *I* is the yearly price increase index; T is duration of the period when important demand fluctuations are observed, days.

Thus, considering social parameters, it is advisable to calculate the drug stock level in pharmacy distribution as follows:

$$H^s_n = (T^s_{q-p} + T^s_c + T^s_b + T^s_r) \times \times (O^s_{j\,cn} + O^s_{juf\,n} + O^s_{j\,sf\,n}). \quad (11)$$

Table 4

Differences between the traditional approach and the approach suggested to drug stock setting in pharmacy distribution

| Parameter | Approaches to drug stock setting in pharmacy distribution | | |
|---|---|---|---|
| | traditional | based on the logistic service personalization | |
| | | taking into account financial parameters | taking into account social parameters |
| Objective | to minimize money costs for building up a stock | to minimize money costs for building up a stock | Appropriate fulfillment of the customer's needs |
| Principles | Cost minimization | Cost minimization | Economical efficiency<br>Social responsibility |
| Priority aspect | Financial | Financial | Social |
| Types of stocks | Transport, current, reserve | Transport, quarantine-prevention, current, buffer, reserve, transport, money | Quarantine-prevention, current, buffer, reserve |
| Conditions of use | – | – | A unique information space for partners within pharmacy logistic and supply chains |
| Methodological framework | Methodology for determining the quota for current assets. – Kyiv : Ministry of Industrial Policy of Ukraine, 1998 [5] | | GDP Requirements [1]<br>GSP Requirements [2] |
| Users | Financial Department | Financial Department | Logistics Department |

The studies have allowed determining differences between the traditional approach and the approach suggested to drug stock setting at the WPC (Tab. 4).

CONCLUSIONS

1. This paper has shown that stock setting in pharmacy distribution needs special attention due to a number of causes.

2. The methodology of stock setting at the WPC must be based on the social responsibility principle requiring distributors to develop personal logistic solutions to meet the customer's needs and guarantee the physical availability of medications, especially of vital drugs. These requirements must be taken into account when setting the stock in pharmacy distribution.

3. In social terms, the drug stock should contain the following groups: quarantine-prevention, current, buffer, reserve.

4. In order to make personal logistic solutions for the customer service and determine the volume of a daily sale for the social component of stock setting in pharmacy distribution, the ABC-VEN analysis has been suggested; it allows to differentiate the drug stock line at the WPC in terms of their importance not only to provide the profitability of the WPC, but also to take into account their importance for customers.

REFERENCES

1. *Лікарські засоби. Належна практика дистрибуції. СТ-Н МОЗУ 42-5.0:2014. – К.: МОЗУ, 2014. – 51 с.*
2. *Лікарські засоби. Належна практика зберігання. СТ-Н МОЗУ 42-5.0:2014. – К.: МОЗУ, 2014. – 23 с.*
3. *Посилкіна О.В. // Управління, економіка та забезпечення якості в фармації. – 2015. – №1 (39). – С. 80-84.*
4. *Славич-Приступа А.С. Практический маркетинг для аптек. – М.: Ремедиум, 2005. – 138 с.*
5. *Methodology for defining the quota for current assets. – К.: Ministry of Industrial Policy of Ukraine, 1998. – 5 с.*

**РОЗРОБКА МЕТОДИЧНИХ ЗАСАД НОРМУВАННЯ ЗАПАСІВ ЛІКАРСЬКИХ ЗАСОБІВ У ФАРМАЦЕВТИЧНІЙ ДИСТРИБУЦІЇ**

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***Ключові слова:*** *індивідуалізація; логістичний сервіс; клієнт; соціальна відповідальність*

*Доведено, що застосування традиційного підходу до нормування запасів ЛЗ у фармацевтичній дистрибуції призводить, скоріше, до визначення рівня іммобілізованих грошових коштів на створення запасів та не враховує потреб, очікувань, особливостей клієнтів та різної соціальної значущості окремих фармакотерапевтичних груп ЛЗ. Методологія, на якій ґрунтується існуючий підхід до нормування запасів, не відповідає сучасним вимогам щодо належного рівня обслуговування клієнтів. Тому виникла необхідність удосконалення підходу до нормування запасів лікарських засобів у фармацевтичній дистрибуції. Соціальна відповідальність фармацевтичного бізнесу передбачає необхідність застосування не тільки економічних, але й соціальних параметрів при нормуванні запасів ЛЗ. З точки зору соціального аспекту завдання нормування запасів полягає у визначенні переліку ЛЗ та їх обсягів, що повинні бути на складі для належного забезпечення потреб клієнтів. З точки зору соціального аспекту запаси повинні розподілятися на такі види: карантинно-підготовчий, поточний, буферний,*

*страховий. Обсяг одноденної реалізації розраховується з врахуванням обсягів реалізації в разі відсутності сезонних коливань з врахуванням тенденції до зміни частоти призначення ЛЗ за нозологією; страхового запасу ЛЗ на випадок непередбаченого коливання попиту; у період сезонного коливання попиту. З метою забезпечення високого рівня надійності обслуговування клієнтів для визначення рівня страхового запасу пропонується застосовувати ABC-VEN аналіз.*

**РАЗРАБОТКА МЕТОДИЧЕСКИХ ОСНОВ НОРМИРОВАНИЯ ЗАПАСОВ ЛЕКАРСТВЕННЫХ СРЕДСТВ В ФАРМАЦЕВТИЧЕСКОЙ ДИСТРИБУЦИИ**

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*Ключевые слова: индивидуализация; логистический сервис; клиент; социальная ответственность*

*Доказано, что применение традиционного подхода к нормированию запасов ЛС в фармацевтической дистрибуции приводит, скорее, к определению уровня иммобилизованных денежных средств на создание запасов и не учитывает потребностей, ожиданий, особенностей клиентов и разной социальной значимости отдельных фармакотерапевтических групп ЛС. Методология, на которой основывается существующий подход к нормированию запасов, не отвечает современным требованиям относительно надлежащего уровня обслуживания клиентов. Поэтому возникла необходимость усовершенствования подхода к нормированию запасов лекарственных средств в фармацевтической дистрибуции. Социальная ответственность фармацевтического бизнеса предусматривает необходимость применения не только экономических, но и социальных параметров при нормировании запасов ЛС. С точки зрения социального аспекта задание нормирования запасов заключается в определении перечня ЛС и их объемов, что должны быть на складе для надлежащего обеспечения потребностей клиентов. С точки зрения социального аспекта запасы должны распределяться на такие виды: карантинно-подготовительный, текущий, буферный, страховой. Объем одноденной реализации рассчитывается с учетом объема реализации: в случае отсутствия сезонных колебаний с учетом тенденции к изменению частоты назначения ЛС за нозологией; страхового запаса ЛС на случай непредвиденного колебания спроса; в период сезонного колебания спроса. С целью обеспечения высокого уровня надежности обслуживания клиентов для определения уровня страхового запаса предлагается применять ABC-VEN анализ.*

# ЕКСПЕРИМЕНТАЛЬНА ФАРМАКОЛОГІЯ

*Recommended by Doctor of Medicine, professor N.I.Filimonova*

UDC 615.281.9

## THE ANTIMICROBIAL ACTIVITY OF SILVER NANOPARTICLES *IN VITRO*

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*Key words: silver nanoparticles; antimicrobial activity; spectrum of bacteria*

*Silver nanoparticles possess a high potential as an antimicrobial substance against a wide spectrum of bacteria, including antibiotic-resistant strains. Antimicrobial properties of silver nanoparticles with 30 nm in diameter synthesized according to the original protocol have been determined in this study. In in vitro study using the serial dilutions method in the solid medium the minimal inhibition concentration (MIC) of silver nanoparticles against such test-strains as Staphylococcus aureus MRSA ATCC 43300, Pseudomonas aeruginosa ATCC 27853, Escherichia coli ATCC 2592, Shigella sonnei, Salmonella typhimurium 144 was equal to 33.46 μg/ml. MIC against B. subtilis ATCC 6633 was 133.8 μg/ml. The antimicrobial activity of silver nanoparticles has been studied on clinical isolates with multiple drug resistance isolated from wounds, urine, endocervical and faucial scrapings in surgical patients with Klebsiella ozaenae 4348, Citrobacter freundii 4369, Escherichia coli 4358, Enterobacter aerogenes 2476, Proteus mirabilis 4363, Staphylococcus aureus 4312 and Pseudomonas aeruginosa 283. The total inhibition of the microorganisms growth under the action of both doses of silver nanoparticles studied – 10 μg and 20 μg has been observed.*

The search for effective antimicrobial substances is an important task of pharmacology nowadays. The appearance of antibiotic-resistant bacterial strains requires the use of antimicrobial agents with principally new properties compared to traditional drugs, which are able to overcome more successfully the resistance of the causative agents of certain diseases. Metal nanoparticles, and especially silver nanoparticles are in the focus of attention of researchers. It is known that silver nanoparticles are characterized by a pronounced antimicrobial activity. Based on them medicines in the form of creams [6, 9], gels [14], as well as such medical products as catheters [20] and dressings [8, 11] have been developed and introduced into practice. At the Pharmacology Department of O.O.Bohomolets National Medical University the research of pharmacological and toxicological properties of different metal nanoparticles (copper, iron, silver) is carried out. In terms of continuing to study antimicrobial properties of nanosilver an experimental substance of silver nanoparticles (AgNP) with the size of 30 nm has been studied.

**Materials and Methods**

Silver nanoparticles were obtained by means of the chemical reduction method in the aqueous medium according to the original protocol developed in F.D. Ovcharenko Institute of Biocolloidal Chemistry of NAS of Ukraine. They were characterized by size using dynamic light scattering (Zetasizer-3, "Malvern Instruments Ltd", Great Britain) and shape using transmission electron microscopy (JEM-1230, "JEOL", Japan).

The evaluation of the antimicrobial activity of silver nanoparticles was carried out in *in vitro* studies using two methods.

1. The antimicrobial activity of AgNP against test-strains of microorganisms was determined according to "MUC 4.2.1890-04, 2004" by the method of serial dilutions in agar and expressed in the concentration of AgNP per volume unit [2]. The initial concentration of the AgNP solution was 800 μg/ml. The following test-strains were used in this study: *Staphylococcus aureus* MRSA ATCC 43300, *Pseudomonas aeruginosa* ATCC 27853, *Escherichia coli* ATCC 2592, *Bacillus subtilis* ATCC 6633 and strains *Shigella sonnei*, *Salmonella typhimurium* 144 obtained from the collection of the State Research Institute of Biotechnology and Strains. The inoculation doses of test-strains were $10^3$, $10^4$ and $10^5$ CFU/cm$^3$. The antimicrobial effectiveness of AgNP was studied in the final concentrations of 133.8 μg/ml, 100.38 μg/ml, 66.9μg/ml, and 33.46 μg/ml in the nutrient medium (Mueler-Hinton agar). A sterile water dispersion of AgNP was introduced into a sterile Mueler-Hinton agar cooled to 50°C, then mixed and poured on Petri dishes. Cultivation of microorganisms was carried out in thermostat at the temperature of 37°C for 24 h.

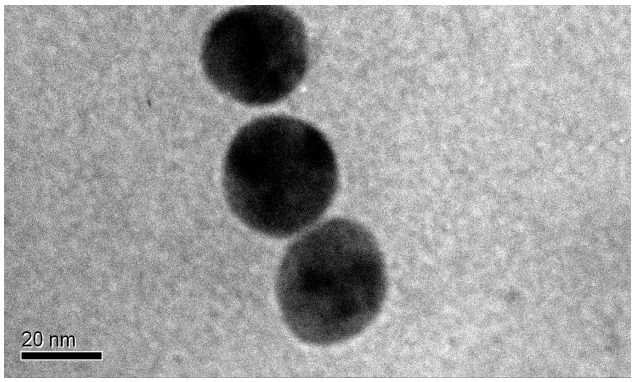



Fig. 1. TEM image of spherical silver nanoparticles of 30 nm in size.

2. The second study was performed on a solid nutrient medium using the method of dosed droplets. Microorganisms involved in the study were multiple drug resistant clinical isolates from wounds, urine, endocervical and faucial scrapings in surgical patients with *Klebsiella ozaenae* 4348, *Citrobacter freundii* 4369, *Escherichia coli* 4358, *Enterobacter aerogenes* 2476, *Proteus mirabilis* 4363, *Staphylococcus aureus* 4312, *Pseudomonas aeruginosa* 283. Microorganisms were inoculated on Mueler-Hinton agar in the concentration of $10^5$ and $10^7$ CFU/cm$^3$ to form a bacterial lawn. Suspensions of microorganisms were prepared using 0.9% saline solution. After 30 min of drying of Petri dishes with inoculated bacterial cultures droplets of aqueous dispersions containing nanoparticles with the concentration of 800 μg/ml by metal were applied onto the agar surface. The droplets were 12.5 and 25 μl in volume and contained 10 and 20 μg of silver nanoparticles, respectively. Bacterial cultures were then cultivated in thermostat for 24 h at the temperature of 37°C. The calculation of results was carried out by measuring the diameter of growth inhibition zones. After measurements of growth inhibition zones Petri dishes were stored for 15 days at the room temperature for detecting any secondary growth.

**Results and Discussion**

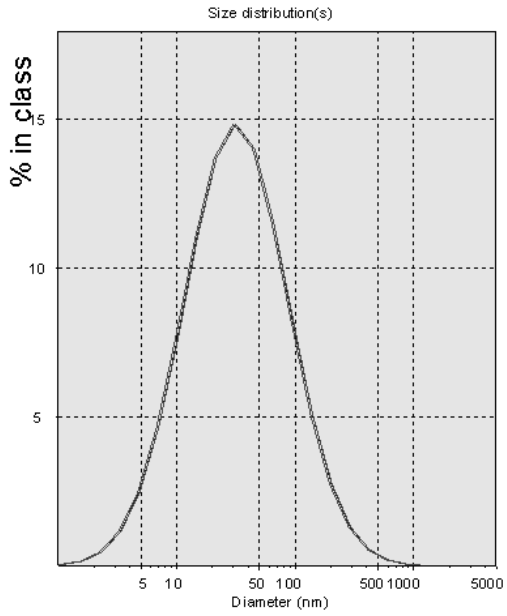
Previous studies proved safety of the AgNP studied, namely genotoxicity, mutagenic action, effect on probiotic bacteria of the gastrointestinal tract [1].

Transmission electron microscopy confirmed that AgNP have a spherical shape (Fig. 1). Results of dynamic light scattering measurements are shown in Fig. 2.

According to the graph it is seen that the size distribution of nanoparticles is monomodal, it means that the colloidal solution has no other fractions. The ZAve parameter shows the average size of nanoparticles. Its average value was 31.8 nm and the absolute error was 0.8 nm.

In Tab. 1 the results of *in vitro* study of the antimicrobial activity of silver nanoparticles against test-strains of microorganisms are given. All strains of microorganisms studied were susceptible to silver nanoparticles. The results obtained indicate a pronounced antimicrobial activity against *Staphylococcus aureus* MRSA ATCC 43300, *Pseudomonas aeruginosa* ATCC 27853, *Escherichia coli* ATCC 2592, *Shigella sonnei*, *Salmonella typhimurium* 144 in the concentration of 33.46 μg/ml. Complete growth inhibition of *Bacillus subtilis* ATCC 6633 was observed at a higher concentration of AgNP – 133.8 μg/ml. This result is important since it is known that *B. subtilis* is a component of the human normal microflora [13]. It is known that bacteria from *Bacillus* genus may develop resistance to silver nitrate and can be used in the process of biological synthesis of silver nanoparticles [10]. The mechanism of appearance of *Bacillus sp.* resistance is uncertain and may be associated with the presence of nitrate reductase enzyme in the bacteria [16].

A pronounced antibacterial activity of AgNPs was also revealed against antibiotic-resistant clinical isolates,




| Run | Angle | KCps. | ZAve | Poly | Fit | Time |
|---|---|---|---|---|---|---|
| 1 | 90.0 | 54.4 | 30.9 | 1.000 | 0.001619 | 13:13:00 |
| 2 | 90.0 | 54.3 | 32.1 | 1.000 | 0.001599 | 13:22:35 |
| 3 | 90.0 | 54.1 | 32.3 | 1.000 | 0.001124 | 13:32:11 |
| Average | | 54.3 | 31.8 | 1.000 | | |
| +/- | | 0.2 | 0.8 | 0.000 | | |

Fig. 2. Dynamic light scattering data of silver nanoparticles. The graph demonstrates distribution of silver nanoparticles by size. The ZAve parameter shows the diameter of nanoparticles , "+/-" – is the absolute error.

Table 1

The antimicrobial activity of 30 nm silver nanoparticles against test-strains of microorganisms

| Test-strains | Inoculation dose of test-strains, CFU/cm$^3$ | The final concentration of AgNP drug in the medium, μg/ml by metal | | | | Reference test-strain growth |
|---|---|---|---|---|---|---|
| | | 133.8 | 100.38 | 66.9 | 33.46 | |
| *Staphylococcus aureus* MRSA ATCC 43300 | $10^3$ | Ø | Ø | Ø | Ø | ++++ |
| | $10^4$ | Ø | Ø | Ø | Ø | ++++ |
| | $10^5$ | Ø | Ø | Ø | Ø | ++++ |
| *Pseudomonas aeruginosa* ATCC 27853 | $10^3$ | Ø | Ø | Ø | Ø | ++++ |
| | $10^4$ | Ø | Ø | Ø | Ø | ++++ |
| | $10^5$ | Ø | Ø | Ø | Ø | ++++ |
| *Escherichia coli* ATCC 2592 | $10^3$ | Ø | Ø | Ø | Ø | ++++ |
| | $10^4$ | Ø | Ø | Ø | Ø | ++++ |
| | $10^5$ | Ø | Ø | Ø | Ø | ++++ |
| Shigella sonnei | $10^3$ | Ø | Ø | Ø | Ø | ++++ |
| | $10^4$ | Ø | Ø | Ø | Ø | ++++ |
| | $10^5$ | Ø | Ø | Ø | Ø | ++++ |
| *Salmonella typhimurium* 144 | $10^3$ | Ø | Ø | Ø | Ø | ++++ |
| | $10^4$ | Ø | Ø | Ø | Ø | ++++ |
| | $10^5$ | Ø | Ø | Ø | Ø | ++++ |
| *B. subtilis* ATCC 6633 | $10^3$ | Ø | Ø | Ø | +++ | +++++ |
| | $10^4$ | Ø | Ø | + | +++++ | +++++ |
| | $10^5$ | Ø | + | +++ | +++++ | +++++ |

Notes: "Ø" – complete inhibition of growth; "++++" – intensive growth; "+++" – weak growth inhibition; "+" – only the growth of single colonies was observed.

and inhibition of bacterial growth was observed in all cases. The results obtained showed that clinical isolates such as *Pseudomonas aeruginosa* 283 and *Klebsiella ozaenae* 4348 which were resistant to the majority of antibiotics, appeared to be susceptible to silver nanoparticles. Diameters of growth inhibition zones for *P. aeruginosa* were the largest among all strains tested. Both gram-negative bacteria from *Enterobacteriaceae* family (*K. ozaenae, E. aerogenes, C. Freundii, E.coli, P. mirabilis*) and gram-positive coccus of *S. aureus* appeared to be susceptible to silver nanoparticles (Tab. 2). Effective concentrations of silver nanoparticles varied from

Table 2

The antimicrobial activity of silver nanoparticles (30 nm) against antibiotic-resistant clinical isolates

| Test-strain | Inoculation dose of test-strain, CFU/cm$^3$ | Diameter* of the growth inhibition zone of clinical isolates under the action of AgNP | |
|---|---|---|---|
| | | 10 μg of AgNP in droplet (by metal) | 20 μg of AgNP in droplet (by metal) |
| *Escherichia coli* 4358 | $10^5$ | 10 | 15 |
| | $10^7$ | 11 | 14 |
| *Klebsiella ozaenae* 4348 | $10^5$ | 12 | 15 |
| | $10^7$ | 12 | 15 |
| *Enterobacter aerogenes* 2476 | $10^5$ | 10 | 13 |
| | $10^7$ | 10 | 12 |
| *Proteus mirabilis* 4363 | $10^5$ | 13 | 17 |
| | $10^7$ | 13 | 17 |
| *Citrobacter freundii* 4369 | $10^5$ | 10 | 15 |
| | $10^7$ | 11 | 14 |
| *Pseudomonas aeruginosa* 283 | $10^5$ | 14 | 20 |
| | $10^7$ | 12 | 20 |
| Staphylococcus aureus 4312 | $10^5$ | 10 | 13 |
| | $10^7$ | 10 | 13 |

Note: * diameters of the growth inhibition zones are expressed in mm.

Table 3

Doses of silver nanoparticles that inhibited growth of antibiotic-resistant strains and expressed in micrograms per surface area of a nutrient medium

| Test-strain | Inoculation dose of test-strain, CFU/cm$^3$ | Doses of silver nanoparticles* | |
|---|---|---|---|
| | | 10 μg of AgNP in droplet (by metal) | 20 μg of AgNP in droplet (by metal) |
| *Escherichia coli* 4358 | $10^5$ | 0.13 | 0.06 |
| | $10^7$ | 0.11 | 0.06 |
| *Klebsiella ozaenae* 4348 | $10^5$ | 0.09 | 0.06 |
| | $10^7$ | 0.09 | 0.06 |
| *Enterobacter aerogenes* 2476 | $10^5$ | 0.13 | 0.08 |
| | $10^7$ | 0.13 | 0.09 |
| *Proteus mirabilis* 4363 | $10^5$ | 0.08 | 0.04 |
| | $10^7$ | 0.08 | 0.04 |
| *Citrobacter freundii* 4369 | $10^5$ | 0.13 | 0.06 |
| | $10^7$ | 0.11 | 0.06 |
| *Pseudomonas aeruginosa* 283 | $10^5$ | 0.06 | 0.03 |
| | $10^7$ | 0.09 | 0.03 |
| Staphylococcus aureus 4312 | $10^5$ | 0.13 | 0.08 |
| | $10^7$ | 0.13 | 0.08 |

Note: * Doses of silver nanoparticles are expressed in μg/mm$^2$.

0.03 μg/mm$^2$ to 0.13 μg/mm$^2$ calculated with reference to the surface area of the nutrient medium (for detailed information see Tab. 3). There was no secondary growth observed in all growth inhibition zones in 15 days of observation.

The mechanism of the antibacterial action of silver nanoparticles is insufficiently studied. The opinion that effect of silver nanoparticles is associated with generation of reactive oxygen species inside the cell is widespread [7, 12, 18, 21]. According to the data [11, 17] a possible mechanism of action of silver nanoparticles includes the complex of the following factors:

- Silver nanoparticles are adsorbed on the surface of the membrane of microorganisms.
- Nanoparticles destroy molecules of lypopolisaccharide and form "sites" of high permeability in the membrane. Silver nanoparticles penetrate inside the cell releasing the $Ag^+$-ions, which cause the following effects:
  - silver ions interact with cytochromes and block the respiratory chain;
  - silver can also interact with DNA inhibiting its replication.

It has been reported that antimicrobial properties of AgNP depend on size and geometry of particles. According to Kahru A., Dubourguier H.-C. [15] the inhibiting action of nanoparticles against nitrifying bacteria was more pronounced if the size of nanoparticles was less than 5 nm. Pal S. et al. [19] have found that there is dependence between the effect of silver nanoparticles and their geometrical parameters. Thus, AgNP with a triangular shape revealed higher activity than spherical shaped nanoparticles. Researchers explain this regularity by high density of silver atoms in triangular nanoparticles, which along with a high specific surface area provide more active interaction with bacterial cells.

CONCLUSIONS

1. The minimal inhibitory concentration of silver nanoparticles against test-strains of such microorganisms as *Staphylococcus aureus* MRSA ATCC 43300, *Pseudomonas aeruginosa* ATCC 27853, *Escherichia coli* ATCC 2592, *Shigella sonnei*, *Salmonella typhimurium* 144 was 33.46 μg/ml, and it was 133.8 μg/ml against *Bacillus subtilis* ATCC 6633.

2. Silver nanoparticles are active against such antibiotic-resistant clinical isolates as *Klebsiella ozaenae* 4348, *Citrobacter freundii* 4369, *Escherichia coli* 4358, *Enterobacter aerogenes* 2476, *Proteus mirabilis* 4363, *Staphylococcus aureus* 4312, *Pseudomonas aeruginosa* 283. Effective concentrations of silver nanoparticles varied from 0.03 μg/mm$^2$ to 0.13 μg/mm$^2$ calculated with reference to the surface area of the nutrient medium.

REFERENCES

1. *Грузіна Т.Г., Дибкова С.М., Прискока А.О. та ін. // Фармакол. та лікарська токсикол. – 2012. – №3. – С. 40-46.*
2. *Методические указания МУК 4.2.1890-04 // Клин. микробиол. антимикроб. химиотер. – 2004. – Т. 6, №4. – С. 306-359.*
3. *Патон Б.Є., Москаленко В.Ф., Чекман І.С., Мовчан Б.О. // Вісник Національної академії наук України. – 2009. – №6. – С. 76-80.*

4. *Чекман І.С. Нанофармакол. – К.: Задруга, 2011. – 424 с.*
5. *Чекман І.С., Симонов П.В. Природні наноструктури та наномеханізми. – К.: Задруга, 2012. – 104 с.*
6. *Чекман І.С., Ульберг З.Р., Маланчук В.О. та ін. Нанонаука, нанобіологія, нанофармація. – К.: Поліграф плюс, 2012. – 328 с.*
7. *Banerjee M., Mallick S., Paul A. et al. // Langmuir. – 2010. – Vol. 26, №8. – P. 5901-5908.*
8. *Bhattacharaya R., Mukherjee P. // Advanced Drug Delivery Rev. – 2008. – Vol. 60. – P. 1289-1306.*
9. *Bhol K.C., Alroy J., Schechter P.J. // Clin. Exp. Dermatol. – 2004. – Vol. 26, №3. – P. 282-287.*
10. *Das V.L., Tomas R., Varghese R.T. et al. // Biotechnol. – 2014. – Vol. 4. – P. 121-126.*
11. *Dunn K. // Burns. – 2004. – Vol. 30. – P. 1-9.*
12. *Gunawan C., Teoh W.Y., Marquis C.P. et al. // Small. – 2013. – Vol. 9, №21. – P. 3554-3560.*
13. *Hong H.A., Khaneja R., Tam N. et al. // Res. in Microbiol. – 2009. – Vol. 160, №2. – P. 134-143.*
14. *Jain J., Arora S., Rajwade J.M. et al. // Mol. Pharm. – 2009. – Vol. 6, №5. – P. 1388-1401.*
15. *Kahru A., Dubourguier H.-C. // Toxicol. – 2010. – Vol. 269. – P. 105-119.*
16. *Kalimuthu K., Babu R.S., Venkataraman D. et al. // Colloids and Surfaces B: Biointerfaces. – 2008. – Vol. 65. – P. 150-153.*
17. *Li Q., Mahendra D., Lyon D. et al. // Water Res. – 2008. – Vol. 42. – P. 4591-4602.*
18. *Nagy A., Harrison A., Sabbani S. et al. // Int. J. Nanomedicine. – 2011. – Vol. 6. – P. 1833-1852.*
19. *Pal S., Tak K.Y., Song J.M. // Applied and Environmental Microbiol. – 2007. – Vol. 73, №6. – P. 1712-1720.*
20. *Roe D., Karandikar B., Bonn-Savage N. et al. // J. of Antimicrobial Chemotherapy. – 2008. – Vol. 61. – P. 869-876.*
21. *Zhang W., Li Y., Niu J., Chen Y. // Langmuir. – 2013. – Vol. 29, №15. – P. 4647-4651.*

**АНТИМІКРОБНА АКТИВНІСТЬ НАНОЧАСТИНОК СРІБЛА *IN VITRO***

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***Ключові слова*:** *наночастинки срібла; антимікробна активність; спектр бактерій*

*Наночастинки срібла мають великий потенціал у якості антимікробного засобу проти широкого спектра бактерій, включаючи антибіотикорезистентні штами. В даному дослідженні визначені антимікробні властивості синтезованих за оригінальним протоколом наночастинок срібла діаметром 30 нм. В дослідах in vitro із використанням методу серійних розведень у твердому середовищі мінімальна інгібуюча концентрація (МІК) по відношенню до тест-штамів Staphylococcus aureus MRSA ATCC 43300, Pseudomonas aeruginosa ATCC 27853, Escherichia coli ATCC 2592, Shigella sonnei, Salmonella typhimurium 144 складала 33,46 мкг/мл. Мінімальна інгібуюча концентрація по відношенню до B. subtilis ATCC 6633 становила 133,8 мкг/мл. Антимікробна активність наночастинок срібла досліджена й на клінічних антибіотикорезистентних ізолятах, що були виділені від хворих хірургічного профілю із ран, сечі, зі скребів з цервікального каналу, зіву: Klebsiella ozaenae 4348, Citrobacter freundii 4369, Escherichia coli 4358, Enterobacter aerogenes 2476, Proteus mirabilis 4363, Staphylococcus aureus 4312 та Pseudomonas aeruginosa 283. Спостерігали повне пригнічення росту досліджуваних клінічних ізолятів при використанні дози наночастинок срібла 10 мг та 20 мг.*

**ПРОТИВОМИКРОБНАЯ АКТИВНОСТЬ НАНОЧАСТИЦ СЕРЕБРА *IN VITRO***

***А.О.Прискока, А.В.Руденко, Л.С.Резниченко, Т.Г.Грузина, З.Р.Ульберг, И.С.Чекман***

***Ключевые слова*:** *наночастицы серебра; противомикробная активность; спектр бактерий*

*Наночастицы серебра имеют большой потенциал в качестве противомикробного средства против широкого спектра бактерий, включая антибиотикорезистентные штаммы. В данном исследовании определены противомикробные свойства синтезированных по оригинальному протоколу наночастиц серебра диаметром 30 нм. В опытах in vitro с использованием метода серийных разведений в твердой среде минимальная ингибирующая концентрация (МИК) в отношении тест-штаммов Staphylococcus aureus MRSA ATCC 43300, Pseudomonas aeruginosa ATCC 27853, Escherichia coli ATCC 2592, Shigella sonnei, Salmonella typhimurium 144 составляла 33,46 мкг/мл. Минимальная ингибирующая концентрация по отношению к Bacillus subtilis ATCC 6633 составляла 133,8 мкг/мл. Противомикробная активность наночастиц серебра исследована на клинических антибиотикорезистентных изолятах, выделенных из ран, мочи, соскобов из цервикального канала, зева больных хирургического профиля: Klebsiella ozaenae 4348, Citrobacter freundii 4369, Escherichia coli 4358, Enterobacter aerogenes 2476, Proteus mirabilis 4363, Staphylococcus aureus 4312 и Pseudomonas aeruginosa 283. Наблюдалось полное подавление роста исследуемых клинических изолятов при использовании дозы наночастиц серебра 10 мг и 20 мг.*

*Recommended by Doctor of Pharmacy, professor V.I.Chuyeshov*

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# A COMPARATIVE STUDY OF EFFICIENCY OF DIFFERENT IVERMECTIN MEDICINAL FORMS FOR TREATING HELMINTOSIS IN LIVE-STOCK ANIMALS

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*Key words: helmintosis; ivermectin; parasites*

*A comparative study of efficiency of injection and oral forms of ivermectin has been conducted. The research results indicate that efficiency of drugs with ivermectin against endoparasites is very high and reaches 100%. The exception is Oesophagostomum dentatum, the therapeutic effectiveness against it in some cases was less – from 85% to 95%. The therapeutic effectiveness in experimental groups is 100%; it confirms the complete parasites elimination by "Neoverm" in the doses from 0.2 to 0.3 mg/kg when using it twice and in the dose of 0.3 mg/kg with a single use. The single use of "Neoverm" in the doses lower than 0.3 mg/kg does not provide the complete elimination of parasites. The use of drugs with ivermectin – "Neoverm" and "Intramek" based on a nanoemulsion provides the therapeutic effect even after single administration. In spite of the fact that a single administration of injection drug "Intramek" in the recommended dose of 0.3 мg/kg did not result in the complete elimination of O. dentatum, its efficiency can be considered as rather sufficient compared to the oral drug "Neoverm". The oral 0.6% ivermectin premix also showed high efficiency against endoparasites, except for O. dentatum. The use of injection and oral drugs with ivermectin– "Neoverm" and "Intramek" based on nanoemulsions is technologically more advantageous for farms than application of the oral premix since these drugs provide a therapeutic effect even after a single administration, while the premix must be given during a week. The therapeutic effectiveness of "Neoverm" and "Intramek" can be considered as equivalent. It is obvious that oral drug "Neoverm" will be beneficial for large livestock and injection "Intramek" – for small farms.*

A wide distribution of helminthoses in agriculture of Ukraine requires improvement of the existing antihelmintic drugs and introduction of the new ones from the scientists of veterinary medicine.

One of the high-effective anthelmintics is ivermectin, which possesses the expressed insectoacaricidic and antihelmintic action [4, 5]. Ivermectin affects glutamate-sensitive chloric channels and γ-aminobutiric acid receptors of parasites.

Owing to development of a new research area – nanotechnology, a unique possibility of creating new medicinal forms based on micelles, liposomes and submicronic emulsions (nanoemulsions) has appeared [9, 11]. Nanoemulsions are also known as miniemulsions, ultradisperse emulsions and submicronic emulsions. Nanoemulsions are emulsions with an average particle diameter from 10 to 1000 nm [6]. The particle size depends on the quantitative composition of the emulsion and the ratio of active and auxiliary components [8]. Particles may exist in the "oil-in-water" and "water-in-oil" forms where the core of the particle is oil or water, respectively [10].

Based on the nanoemulsion of ivermectin the Ukrainian researchers created "Neoverm" and "Intramek" drugs for oral and parenteral use that are easy-to-use, non-toxic in therapeutic doses and effective against endo-and ectoparasites [3].

The aim of the study was to conduct a comparative study of the therapeutic effectiveness of injection and oral drugs with ivermectin based on nanoemulsions, as well as to determine the minimal effective dose and the required dosage frequency of "Neoverm".

**Materials and Methods**

The clinical studies of drugs with ivermectin were conducted at "Agroprodservice" agro-enterprise in the Ternopol region. Three drugs were selected for comparative studies:

- "Neoverm" – a nanoemulsion for oral administration produced by "AT Biopharm" (Kharkiv, Ukraine);
- "Intramek" – a nanoemulsion for injections produced by "AT Biopharm" (Kharkiv, Ukraine);
- "Ivomec" – 0.6% oral ivermectin premix produced by Merial (USA) [7].

The clinical studies of drugs were conducted in the following directions:

- qualitative and quantitative study of helminthic invasion in pigs;
- study of the therapeutic effect when using drugs on animals;
- determination of the minimal effective dose and the required dosage frequency of "Neoverm".

The coproovoscopic examination of feces was performed by the method of Fulleborn. Counting the number of helminthic eggs in 1 g of feces was performed by Trach method. Feces taken from all animals were examined for the presence of eggs of pulmonary and gastrointestinal nematodes, the intensity of infection was determined by Trach method [1, 2].

Table 1

The scheme of the experiment

| Group No. | The number of animals in the group | The drug used | Dose and drug regimen |
|---|---|---|---|
| 1 | 20 | no (control) | no |
| 2 | 20 | Neoverm | 0.3 mg AS/1 kg b.w., twice, with 24 h interval |
| 3 | 20 | | 0.25 mg AS/1 kg b.w., twice, with 24 h interval |
| 4 | 20 | | 0.2 mg AS/1 kg b.w., twice, with 24 h interval |
| 5 | 20 | | 0.3 mg AS/1 kg b.w., once |
| 6 | 20 | | 0.25 mg AS/1 kg b.w., once |
| 7 | 20 | | 0.2 mg AS/1 kg b.w., once |
| 8 | 20 | 0.6% Ivomec premix | 0.1 mg AS/1 kg b.w., daily, for 7 days |
| 9 | 20 | Intramek | 0.3 mg AS/1 kg b.w., once |

Table 2

Invasion intensivity (II) and extensivity (IE) values before treatment

| Parasites | IE, % | II, eggs per 1g of feces |
|---|---|---|
| Ascaris suum | 90 | 10-40 |
| Trichuris suis | 45 | 5-20 |
| Oesophagostomum dentatum | 80 | 5-10 |
| Metastrongylus spp. | 21 | 5-10 |
| Strongyloides ransomi | 90 | 5-15 |

Table 3

IE values after treatment, groups 2-7 ("Neoverm")

| Group No. | Parasites | IE, % |
|---|---|---|
| 2 | Ascaris suum | 100 |
| | Trichuris suis | 100 |
| | Oesophagostomum dentatum | 100 |
| | Metastrongylus spp. | 100 |
| | Strongyloides ransomi | 100 |
| 3 | Ascaris suum | 100 |
| | Trichuris suis | 100 |
| | Oesophagostomum dentatum | 100 |
| | Metastrongylus spp. | 100 |
| | Strongyloides ransomi | 100 |
| 4 | Ascaris suum | 100 |
| | Trichuris suis | 100 |
| | Oesophagostomum dentatum | 100 |
| | Metastrongylus spp. | 100 |
| | Strongyloides ransomi | 100 |
| 5 | Ascaris suum | 100 |
| | Trichuris suis | 100 |
| | Oesophagostomum dentatum | 100 |
| | Metastrongylus spp. | 100 |
| | Strongyloides ransomi | 100 |
| 6 | Ascaris suum | 100 |
| | Trichuris suis | 90 |
| | Oesophagostomum dentatum | 90 |
| | Metastrongylus spp. | 100 |
| | Strongyloides ransomi | 100 |
| 7 | Ascaris suum | 100 |
| | Trichuris suis | 90 |
| | Oesophagostomum dentatum | 85 |
| | Metastrongylus spp. | 100 |
| | Strongyloides ransomi | 100 |

Table 4

IE values after treatment, group 9 ("Intramek")

| Group No. | Parasites | IE, % |
|---|---|---|
| 9 | Ascaris suum | 100 |
| | Trichuris suis | 100 |
| | Oesophagostomum dentatum | 95 |
| | Metastrongylus spp. | 100 |
| | Strongyloides ransomi | 100 |

Table 5

IE values after treatment, group 8 (0.6% premix)

| Group No. | Parasites | IE, % |
|---|---|---|
| 8 | Ascaris suum | 100 |
| | Trichuris suis | 100 |
| | Oesophagostomum dentatum | 95 |
| | Metastrongylus spp. | 100 |
| | Strongyloides ransomi | 100 |

The studies for the presence and intensity of invasion were performed in 14 days after treatment.

For the research there were 9 groups of pigs, 20 animals in each group, 5 months old with the body weight of 70-80 kg. Group 1 was the control and received no drugs. The experimental groups 2-9 received drugs with ivermectin in an amount of 0.1-0.3 mg of the active substance (AS) per 1 kg of the body weight (Tab. 1).

**Results and Discussion**

The results of the qualitative and quantitative study of helminthic invasion are given in Tab. 2. *Ascaris suum, Trichuris suis, Oesophagostomum dentatum, Metastrongylus spp., Strongyloides ransomi* were detected. The invasion intensivity was from 5 to 40 eggs per 1 g of feces.

The results of fecal test coproovoscopy in 14 days after treatment are given in Tab. 3-5.

The research results indicate that efficiency of drugs with ivermectin against endoparasites is very high – up to 100%. The exception is *Oesophagostomum dentatum*, the therapeutic effectiveness against it in some cases was less – from 85% to 95% (Tab. 3-5).

The therapeutic effectiveness of drugs in experimental groups 2-5 is 100%; it indicates the complete para-

sites elimination by "Neoverm" in the doses from 0.2 to 0.3 mg/kg when using it twice and in the dose of 0.3 mg/kg with a single use. The single use of "Neoverm" in the doses lower than 0.3 mg/kg does not provide the complete elimination of parasites (Tab. 3).

In spite of the fact that a single administration of injection drug "Intramek" in the recommended dose of 0.3 мg/kg did not result in the complete elimination of *O. dentatum* (Tab. 4), its efficiency can be considered as rather sufficient compared to the oral drug "Neoverm".

The oral 0.6% ivermectin premix also showed high efficiency against endoparasites, except for *O. dentatum* (Tab. 5).

CONCLUSIONS

The use of injection and oral drugs with ivermectin– "Neoverm" and "Intramek" based on nanoemulsions is technologically more advantageous for farms than application of the oral premix since these drugs provide a therapeutic effect even after a single administration, while the premix must be given during a week [7].

A simple calculation shows that when using the oral premix for the period recommended by the manufacturer (7 days) each animal receives 0.7 mg of the active substance per 1 kg of the body weight, and while using "Neoverm" and "Intramek" only 0.3 mg is administered. It significantly decreases the likelihood of penetration of residual quantities of the active substance in a human food and reduces the cost for treatment of animals.

The therapeutic effectiveness of "Neoverm" and "Intramek" can be considered as equivalent. It is obvious that oral drug "Neoverm" will be beneficial for large livestock and injection "Intramek" – for small farms.

REFERENCES


1. *Галат В.Ф., Березовський А.В., Прус М.П., Сорока Н.М. Паразитологія та інвазійні хвороби тварин. – К.: Урожай, 2009. – 363 с.*
2. *Кузьмин А.А. Антгельминтики в ветеринарной медицине. – К.: Аквариум Лтд, 2004. – 144 с.*
3. *Соколов Ю.В., Хасбауи Х.А. // Сучасне птахівництво. – 2014. – №3 (136). – С. 18-20.*
4. *Campbell W.C. // Parasitol. Today. – 1990. – Vol. 6. – P. 130-133.*
5. *Campbell W.C. // New York, Berlin, Heidelberg, London, Paris, Tokyo. – 1989. – P. 1-25, 244-259.*
6. *Fryd M.M., Mason T.G. // Annu. Rev. Phys. Chem. – 2012. – Vol. 63. – P. 493-518.*
7. *IVOMEC PREMIX FOR SWINE 0.6% // Compendium of Veterinary Products. – [Електронний ресурс]. – Спосіб доступу: http://merialce.naccvp.com/product/view/1182040 – Заголовок з екрана.*
8. *Lovelyn C., Attama A.A. // J. of Biomaterials and Nanobiotechnol. – 2011. – Vol. 2. – P. 626-639.*
9. *Salata O. // J. Nanobiotechnol. – 2004. – Vol. 2. – P. 3.*
10. *Shah P., Bhalodia D., Shelat P. // Systematic Rev. in Pharmacy. – 2010. – Vol. 1, №1. – P. 24-32.*
11. *Speiser P.P. // Methods Find. Exp. Clin. Pharmacol. –1991. – №13. – P. 337-342.*


**ПОРІВНЯЛЬНЕ ДОСЛІДЖЕННЯ ЕФЕКТИВНОСТІ РІЗНИХ ЛІКАРСЬКИХ ФОРМ ІВЕРМЕКТИНУ ПРИ ЛІКУВАННІ ГЕЛЬМІНТОЗІВ СІЛЬСЬКОГОСПОДАРСЬКИХ ТВАРИН**

***Ю.В.Соколов***

***Ключові слова:*** *гельмінтоз; івермектин; паразити; екстенсефективність*

*Проведені порівняльні дослідження ефективності ін'єкційної і пероральної форм івермектину. Їх результати свідчать, що ефективність препаратів івермектину проти ендопаразитів є дуже високою – 100%. Виключенням є Oesophagostomum dentatum. Екстенсефективність дії препаратів проти цього виду паразитів в деяких випадках є дещо меншою – від 85% до 95%. Екстенсефективність препаратів у дослідних групах складає 100%, що свідчить про повне знищення паразитів препаратом «Неоверм» в дозах від 0,2 до 0,3 мг/кг при дворазовому застосуванні і в дозі 0,3 мг/кг – при одноразовому. Одноразове застосування «Неоверму» в дозах менше 0,3 мг/кг не забезпечує знищення 100% паразитів. Недивлячись на те, що одноразове застосування ін´єкційного препарату «Інтрамек» в рекомендованій дозі 0,3 мг/кг не призвело до повного знищення O. dentatum, його ефективність можна вважати цілком достатньою у порівнянні з пероральним препаратом «Неоверм». Пероральний 0,6%-вий премікс івермектину також показав високу ефективність проти ендопаразитів, за виключенням O. dentatum. Використання ін'єкційних і пероральних препаратів івермектину на основі наноемульсій «Неоверму» та «Інтрамеку» технологічно більш вигідне для господарств, ніж застосування перорального премікcу, т. я. дані препарати забезпечують терапевтичний ефект навіть після одноразового застосування, а премікс необхідно давати впродовж тижня. Терапевтичну ефективність «Неоверму» та «Інтрамеку» можна вважати еквівалентною. Очевидно, що пероральний препарат «Неоверм» буде вигідний на великому поголів´ї, а ін´єкційний «Інтрамек» – для невеликих господарств.*

**СРАВНИТЕЛЬНОЕ ИССЛЕДОВАНИЕ ЭФФЕКТИВНОСТИ РАЗЛИЧНЫХ ЛЕКАРСТВЕННЫХ ФОРМ ИВЕРМЕКТИНА ПРИ ЛЕЧЕНИИ ГЕЛЬМИНТОЗОВ СЕЛЬСКОХОЗЯЙСТВЕННЫХ ЖИВОТНЫХ**

***Ю.В.Соколов***

***Ключевые слова:*** *гельминтоз; ивермектин; паразиты; экстенсэффективность*

*Проведено сравнительное исследование эффективности инъекционной и пероральной форм ивермектина. Результаты исследования свидетельствуют, что эффективность препаратов ивермектина против эндопаразитов очень высока – 100%. Исключением является Oesophagostomum dentatum. Экстенсэффективность действия препаратов против этого вида паразитов в некоторых случаях несколько меньше – от 85% до 95%. Экстенсэффективность препаратов в опытных группах составляет 100%, что свидетельствует о полном уничтожении паразитов препаратом «Неоверм» в дозах от 0,2 до 0,3 мг/кг при двухкратном применении и в дозе 0,3 мг/кг – при однократном. Однократное использование «Неоверма» в дозах меньше 0,3 мг/кг не обеспечивает уничтожение 100% паразитов. Несмотря на то, что однократное применение инъекционного препарата «Интрамек» в рекомендованной дозе 0,3 мг/кг не привело к полному уничтожению O. dentatum, эффективность препарата можно считать вполне достаточной по сравнению с пероральным препаратом «Неоверм». Пероральный 0,6%-ный премикс ивермектина также показал высокую эффективность против эндопаразитов, за исключением O. dentatum. Применение инъекционных и пероральных препаратов ивермектина на основе наноэмульсий «Неоверм» и «Интрамек» технологически более выгодно для хозяйств, чем использование перорального премикса, т. к. данные препараты обеспечивают терапевтический эффект даже после однократного применения, а премикс необходимо давать на протяжении недели. Терапевтическую эффективность «Неоверма» и «Интрамека» можно считать эквивалентной. Очевидно, что пероральный препарат «Неоверм» будет выгоден на большом поголовье, а инъекционный «Интрамек» – для небольших хозяйств.*

*Recommended by Doctor of Medicine, Professor T.I.Tiupka*

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# THE EFFECT OF IVABRADINE AND ω-3 POLYUNSATURATED FATTY ACIDS ON THE FIBRONECTIN PLASMA LEVELS IN PATIENTS WITH HEART FAILURE

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*Key words: heart failure; treatment; ivabradine; ω-3 polyunsaturated fatty acids*

*Chronic heart failure (HF) is a highly prevalent chronic pathological condition with high morbidity and mortality rates. The cost for HF treatment is 2% of the national health expenditures each year. Fibronectin (Fn) has a crucial role in the process of tissue-specific morphogenesis and cell differentiation in embryogenesis; it stimulates fibroblasts and different growth factors during wound healing and tissue reparation. The aim of our study was to assess the possible effects of Ivabradine and ω-3 polyunsaturated fatty acids on the fibronectin plasma level in patients with heart failure. 357 Patients with ischemic HF and the sinus rhythm were observed. In accordance to treatment regimens all patients were divided into four groups: group I – basic treatment; group II – basic treatment and Ivabradine; group III – basic treatment and PUFA; group IV – basic treatment with Ivabradine and PUFA. The fibronectin levels were measured by ELISA method. The average value of the Fn concentration in HF patients was 1.25 times higher than in the control group ($p<0.05$). In patients with basic HF treatment the tendency to the Fn plasma level decrease ($p>0.05$) was observed. In the group with additional prescription of Ivabradine the concentration of this protein in the blood decreased by 14.5% ($p<0.01$). The same situation was observed in the group with additional use of PUFA – by 11.1% ($p<0.05$). In group IV the plasma concentration of Fn decreased by 12.9% ($p<0.05$). Thus, Ivabradine and ω-3 polyunsaturated fatty acids, either alone or in combination, decrease the plasma levels of fibronectin in patients with heart failure, and it shows a pronounced cardioprotective effect.*

Chronic heart failure (HF) is a highly prevalent chronic pathological condition with high morbidity and mortality rates. The prevalence of HF is 1% to 2% in the general population worldwide and at least 10-20% among the age group of 75 years and older [7]. The cost for HF treatment is 2% each year [11].

Fibronectin (Fn) has a crucial role in the process of tissue-specific morphogenesis and cell differentiation in embryogenesis; it stimulates fibroblasts and different growth factors during wound healing and tissue reparation [10]. The experimental investigations conducted demonstrated the importance of this protein in HF progression [4].

According to the current recommendations the main aim of treatment of HF patients is reduction of symptoms and signs, prevention of hospital admissions and increased life expectancy [5, 7].

Heart rate is an important factor of pathogenesis of coronary artery disease and HF. It is a modifying risk factor [6]. There are a lot of mechanisms of increase of a cardiovascular risk due to the high heart rate: oxygen demand, acceleration of atherosclerosis, and possibility of atheroma rupture [3]. Thus, the heart rate reduction is one of the priority targets in CAD and HF treatment. Ivabradine is a representative of a new class of medicines with reduction effects on the heart rate. It is approved for HF management. There are no data of influence of this medicine on some components of the extracellular matrix.

Taking into account the role of inflammation in HF pathogenesis, ω-3 polyunsaturated fatty acids (ω-3 PUFA) are referred to the drugs used for its treatment. These acids affect the lipid metabolism and various factors of the inflammatory process, thus improving the prognosis of patients with cardiovascular diseases.

The aim of our study was to assess the possible effects of Ivabradine and ω-3 polyunsaturated fatty acids on the fibronectin plasma level in patients with heart failure.

**Materials and Methods**

357 Patients with ischemic HF and the sinus rhythm were observed. In accordance to treatment regimens all patients were randomized as follows: group I (89 patients) – basic treatment; group II (91 patients) – basic treatment and Ivabradine (Coraxan, Les Laboratoires Servier Industrie, France) – 5 or 7.5 mg twice a day; group III (90 patients) – basic treatment and PUFA (Omacor, Abbott Laboratories GmbH, USA) – 1000 mg per day; group IV (87 patients) – basic treatment with Ivabradibe and PUFA in similar doses. All patients were examined before and after 6 months of treatment. The control group was 30 practically healthy persons. The study was performed in accordance with the Helsinki Declaration and Good Clinical Practice Guideline. The study was approved by the local ethics committee, and the written informed consent was obtained from all patients.

The fibronectin levels were measured by ELISA method with "Fibronectin ELISA Kit" (Technoclone GmbH, Austria).

Statistical analysis was performed using a "Statistica for Windows 12.0" programme (StatSoft, Tulsa, OK, USA). Difference was considered significant at $p<0.05$.

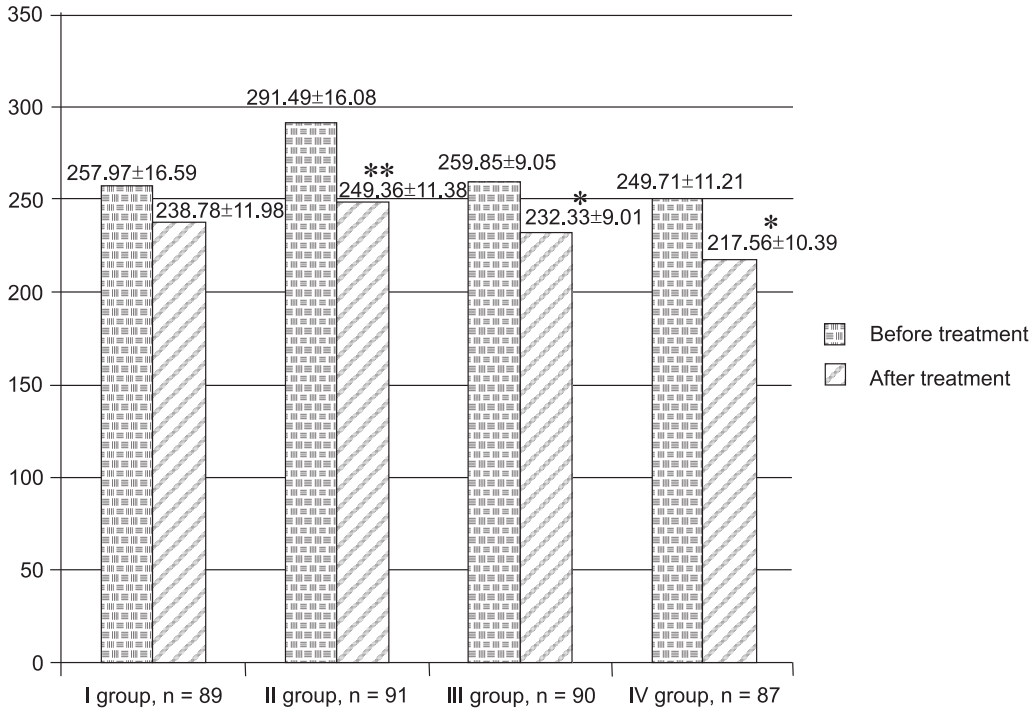


Fig. The dynamics of plasma levels of fibronectin in patients with HF (the probability of the difference between the values before and after treatment: * – p<0.05; ** – p<0.01).

**Results and Discussion**

The average age of the patients with HF examined was (67.98±12.06) years. The average value of the Fn concentration in HF patients was 1.25 times higher than the same value in the control group: (259.55±7.88) μg/mL versus (207.56±13.62) μg/mL (p<0.05).

The Fn plasma levels elevation is one of the factors of atherosclerosis progression. This protein can be compared with a double-edged sword, which on the good edge, promotes the formation of a thick fibrous cap, but on the bad edge, expands the ECM, resulting in more atherogenic lipoprotein retention. This unfavourable consequence alone would result in infiltration of inflammatory cells and plaque progression [9].

In patients with basic HF treatment only tendency to the Fn plasma level decrease was observed (Fig.): from (257.97±16.59) μg/mL to (238.78±11.98) μg/mL (p>0.05). In the group with additional prescription of Ivabradine the concentration of this protein in the blood decreased by 14.5%: from (291.49±16.08) μg/mL to (249.36±11.38) μg/mL (p<0.01). The same decrease of the Fn plasma level was observed in the group with additional use of PUFA – from (259.85±9.05) μg/mL to (232.33±9.01) μg/mL (for 11.1%; p<0.05).

In group IV the plasma concentration of Fn decreased by 12.9%: from (249.71±11.21) μg/mL to (217.56±±10.39) μg/mL (p<0.05).

Some trials showed the influence of activated renin-angiotensin-aldosterone system (RAAS) on elevation of Fn in plasma [1]. In our opinion, the use of RAAS blockers as components of HF management causes a decreasing trend in this parameter.

The effect of ω-3 PUFA on the Fn levels probably occurs via its anti-inflammatory mechanisms. Some researchers suppose that Fn is a protein of the acute phase of inflammation, which takes an active part in the processes of nonspecific immune response [12].

The mechanism of the effect of Ivabradine on the Fn levels reduction in the blood remains unclear. It is obvious that it occurs by two main ways: first – via the heart rate reduction (which is an important factor of myocardial fibrosis [8]); second – via anti-inflammatory effects of Ivabradine (some experimental results showed the decrease of the concentration of interleukin-6, tumour necrosis factor – α and protein of chemotaxis of macrophages-1in the myocardium [2]).

CONCLUSIONS

Ivabradine and ω-3 polyunsaturated fatty acids, either alone or in combination, decrease the plasma levels of fibronectin in patients with heart failure, and it shows a pronounced cardioprotective effect. A promising direction for future research is the study of molecular mechanisms of the drug effect on the processes of fibrosis.

REFERENCES

1. *Сидорчук Л.П. // Укр. терапевт. журн. – 2007. – №3. – С. 33-39.*
2. *Chun L., Teng Z., Na-Dan Z. et al. // PLOS One. – 2012. – Vol. 7 (6). – P. 1-13.*
3. *Deedwania P. // Drugs. – 2013. – Vol. 73. – P. 1569-1586.*
4. *Ding L., Guo D., Homandberg G.A. // Osteoarthritis Cartilage. – 2009. – Vol. 17. – P. 1385-1392.*

5. *Ekman I., Cleland J.G., Andersson B. et al. // Eur. J. Heart Fail. – 2005. – Vol. 7. – P. 699-703.*
6. *Fox K., Ford I., Steg P.G. et al. // Lancet. – 2008. – Vol. 372 (9641). – P. 817-821.*
7. *McMurrey J., Adamopoulus S., Anker S. et al. // Eur. Heart J. – 2012. – Vol. 33. – P. 1787-1847.*
8. *Mewton N., Liu C., Croisillie P. et al. // JACC. – 2011. – Vol. 57. – P. 891-903.*
9. *Moore K.J., Fisher E.A. // EMBO Mol. Med. – 2012. – Vol. 4. – P. 561-563.*
10. *Schultz G.S., Wysocki A. // Wound Repair Regen. – 2009. – Vol. 17. – P. 153-162.*
11. *Stewart S., Jenkins A., Buchan S. et al. // Eur. Heart Fail. – 2002. – Vol. 4. – P. 361-371.*
12. *White E.S., Baralle E.F., Muro A.F. // J. Pathol. – 2008. – Vol. 216. – P. 1-14.*

**ВПЛИВ ІВАБРАДИНУ ТА ω-3 ПОЛІНЕНАСИЧЕНИХ ЖИРНИХ КИСЛОТ НА РІВНІ ПЛАЗМОВОГО ФІБРОНЕКТИНУ У ХВОРИХ ІЗ СЕРЦЕВОЮ НЕДОСТАТНІСТЮ**

***С.В.Федоров***

***Ключові слова:*** *серцева недостатність; лікування; івабрадин; ω-3 поліненасичені жирні кислоти*

*Серцева недостатність (СН) – кінцева стадія більшості захворювань серця та головна причина захворюваності і смертності. На лікування СН витрачається понад 2% національних видатків на охорону здоров'я. Фібронектин (Фн) відіграє ключову роль у процесах тканинно-специфічного морфогенезу та диференціації клітин під час ембріогенезу; стимулює фібробласти та різноманітні ростові фактори під час загоєння ран та репарації тканин. Досліджено ймовірний вплив івабрадину та препарату ω-3 поліненасичених жирних кислот (ω-3 ПНЖК) на вміст фібронектину в плазмі крові хворих із серцевою недостатністю. Обстежені 357 пацієнтів із СН ішемічного ґенезу та зі збереженим синусовим ритмом. За лікувальними схемами хворі були розділені на 4 групи: базова терапія; базова терапія та івабрадин; базова терапія та ω-3 ПНЖК; ω-3 ПНЖК та поєднання обох препаратів. Рівень Фн у плазмі вивчали методом ІФА. Середнє значення концентрації Фн у плазмі крові пацієнтів із СН в 1,25 рази перевищувало подібний показник у контрольній групі. У групі обстежених хворих із базовим лікуванням відмічено тенденцію до зниження показника плазмового Фн ($p>0,05$). Водночас у пацієнтів, яким додатково призначали івабрадин, рівень даного протеїну знизився на 14,5% ($p<0,01$). Подібна картина відмічена також і в групі пацієнтів, які вживали препарат ω-3 ПНЖК в додаток до базового лікування (на 11,1%; $p<0,05$). Лікувальна схема з застосуванням поєднання івабрадину та ω-3 ПНЖК призводила до зменшення концентрації Фн у плазмі крові на 12,9% ($p<0,05$). Таким чином, івабрадин та препарати ω-3 поліненасичених жирних кислот як самостійно, так і в комбінації зменшують рівні фібронектину в плазмі крові хворих із серцевою недостатністю, що є свідченням вираженого кардіопротективного ефекту.*

**ВЛИЯНИЕ ИВАБРАДИНА И ω-3 ПОЛИНЕНАСЫЩЕННЫХ ЖИРНЫХ КИСЛОТ НА УРОВНИ ПЛАЗМЕННОГО ФИБРОНЕКТИНА У БОЛЬНЫХ С СЕРДЕЧНОЙ НЕДОСТАТОЧНОСТЬЮ**

***С.В.Федоров***

***Ключевые слова:*** *сердечная недостаточность; лечение; ивабрадин; ω-3 полиненасыщенные жирные кислоты*

*Сердечная недостаточность (СН) – конечная стадия большинства заболеваний сердца и главная причина заболеваемости и смертности. Расходы на лечение СН составляют 2% национальных расходов на здравоохранение. Фибронектин (Фн) играет важную роль в процессах тканево-специфического морфогенеза и дифференцировки клеток во время эмбриогенеза; стимулирует фибробласты и разные ростовые факторы во время заживления ран и репарации тканей. Изучено возможное влияние ивабрадина и препарата ω-3 полиненасыщенных жирных кислот (ω-3 ПНЖК) на содержание Фн в плазме крови больных с сердечной недостаточностью. Обследованы 357 пациентов с СН ишемического генеза и с сохраненным синусовым ритмом. По лечебным схемам больных разделили на 4 группы: базовое лечение; базовое лечение и ивабрадин; базовое лечение и ω-3 ПНЖК; ω-3 ПНЖК и сочетание двух препаратов. Уровень Фн в плазме определяли методом ИФА. Среднее значение концентрации Фн в плазме крови больных с СН было в 1,25 раза выше аналогичного показателя в группе контроля. В группе базовой терапии отмечена тенденция к снижению показателя плазменного Фн ($p>0,05$). В то же время у пациентов, которым дополнительно назначали ивабрадин, уровень данного протеина снизился на 14,5% ($p<0,01$). Похожая ситуация отмечена в группе больных, которые принимали препарат ω-3 ПНЖК – на 11,1% ($p<0,05$). Лечебная схема с использованием ивабрадина и ω-3 ПНЖК уменьшала концентрацию Фн в плазме на 12,9% ($p<0,05$). Таким образом, ивабрадин и препараты ω-3 полиненасыщенных жирных кислот как самостоятельно, так и в комбинации, уменьшают уровни фибронектина в плазме крови больных с сердечной недостаточностью, что свидетельствует о выраженном кардиопротекторном эффекте.*

*Recommended by Doctor of Pharmacy, professor L.S.Strelnikov*

UDC 615.281:577.112.34:547.466: 547.233.1/.4

# THE EFFECT OF SUBSTITUENTS IN THE MOLECULES OF N-, R-ALKYL AMINES ON SOME GRAM-POSITIVE STRAINS OF MICROORGANISMS

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*Key words: N-, R-alkylamines; antimicrobial activity*

*The intensive use of antibiotics in patient care institutions often without determining specificity and sensitivity to them leads to more rapid development of resistance to pathogens of nosocomial infections; that is why it is one of the urgent problems of healthcare of Ukraine. The most common gram-positive pathogens of nosocomial infections are Staphylococcus aureus, coagulase-negative staphylococci and enterococci. In order to determine dependence of the microbiological action on the nature of substituents in the molecules of N-, R-alkylamines some methyl, ethylamine, aminoalcohols, N-hydroxymethyl- and N-methyl-N-carboxymethylamines have been tested. In accordance with the WHO recommendations to assess the antibacterial activity of N-, R-alkylamines the gram-positive test strains – Staphylococcus aureus ATCC 25923 and Bacillus subtilis ATCC 6633 were used. The aliphatic amines and aminoalcohols studied show a weak or moderate activity in relation to strains of Staphylococcus aureus ATCC 25923 and Bacillus subtilis ATCC 6633. Compounds containing a carboxyl group and methyl radicals in the molecule exhibit the greatest antimicrobial activity in relation to the gram-positive strains of microorganisms under research.*

Antibacterial agents are practically the only group of drugs which effectiveness decreases with time due to development of resistance. The intensive use of antibiotics in patient care institutions often without determining specificity and sensitivity to them leads to more rapid development of resistance to pathogens of nosocomial infections; that is why it is one of the urgent problems of healthcare of Ukraine. The main factors that contribute to the increased disease incidence are: shortage of medicines, antiseptics, detergents and disinfectants, medical instruments, linen, sterilization equipment since medical preventive institutions are forced to work in conditions of the extremely limited funding; a significant growth in the number of hospital strains that are resistant to antibiotics and disinfectants, etc. [4].

The most common gram-positive pathogens of nosocomial infections are *Staphylococcus aureus*, coagulase-negative staphylococci and enterococci. The results of the multicentre randomized trial SCOPE (USA) published in 2004 indicate the predominance of gram-positive cocci in the etiological structure of nosocomial bacteriemias [14].

This tendency creates significant problems since the choice of antimicrobial agents intended to combat drug-resistant gram-positive microorganisms is limited. It should be also noted the fact that antibiotics are less than 5% of the drugs being currently at the stage of drug development [5], and as resistance to the medicines used develops, there is a need in both creation of new drugs, and correction of methods for using the existing ones. Therefore, the research in development of drugs, including vaccines and diagnostic agents, are of vital importance for protection of future generations.

It is known that tertiary amine salts or quaternary ammonium bases containing radicals with a large number of carbon atoms exhibit a strong bacteriostatic and bactericidal action, as well as possess pronounced disinfectant properties [1, 8]. In literature there are data that compounds with the number of carbon atoms from 5 to 16 are the most effective against microorganisms [6].

The aim of this work was to determine the effect of various functional groups containing in the molecules of N-, R-alkylamines derivatives on their antibacterial activity in relation to some gram-positive strains of microorganisms.

**Materials and Methods**

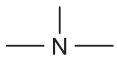
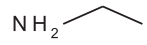
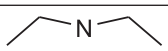
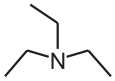
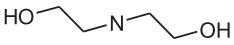
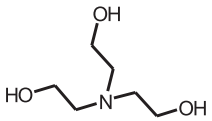
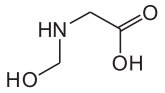
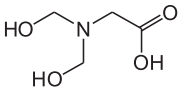
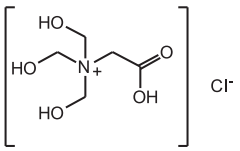
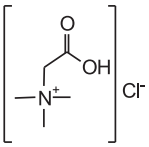
In order to determine dependence of the microbiological action on the nature of substituents in the molecules of N-, R-alkylamines the following groups were tested: alkylamines (compounds I-VI), aminoalcohols (compounds VII-IX), N-hydroxymethyl-N-carboxymethylamines (compounds X-XII), and N-methyl-N-carboxymethylamines (compounds XIII-XV) (see Table).

The compounds under research were obtained from commercial sources or synthesized according to the synthetic schemes previously developed [2, 3, 6, 9, 11-13]. Reagents were purchased from "Sigma-Aldrich" (USA) and used without further purification. 1% Aqueous solutions of compounds I-XV were tested.

In accordance with the WHO recommendations to assess the antibacterial activity of N-, R-alkylamines the gram-positive test strains – *Staphylococcus aureus*

Table

Data of the study results of the antibacterial activity of 1% solutions of the samples under research

| Compound | The structural formula | Test strains/inhibition zones, mm | |
|---|---|---|---|
| | | Staphylococcus aureus ATCC 25923 | Bacillus subtilis ATCC 6633 |
| I |  | 10 (l.s.)* ± 0.1 | 10 (l.s.) ± 0.1 |
| II |  | 10 (l.s.) ± 0.1 | 10 (l.s.) ± 0.1 |
| III |  | 15 (l.s.) ± 0.1 | 15 (l.s.) ± 0.1 |
| IV |  | 10 (l.s.) ± 0.1 | 10 (l.s.) ± 0.1 |
| V |  | 12 (l.s.) ± 0.1 | 10 (l.s.) ± 0.1 |
| VI |  | 10 (l.s.) ± 0.1 | 10 (l.s.) ± 0.1 |
| VII |  | 10 (l.s.) ± 0.1 | 10 (l.s.) ± 0.1 |
| VIII |  | 13 (l.s.) ± 0.1 | 10 (l.s.) ± 0.1 |
| IX |  | 20 (s.) ± 0.2 | 21 (s.) ± 0.2 |
| X |  | 20 (s.) ± 0.2 | 17 (s.) ± 0.2 |
| XI |  | 20 (s.) ± 0.2 | 25 (s.) ± 0.2 |
| XII |  | 21 (s.) ± 0.2 | 25 (s.) ± 0.2 |
| XIII |  | 40 (h.s.) ± 0.3 | 39 (h.s.) ± 0.3 |
| XIV |  | 40 (h.s.) ± 0.3 | 39 (h.s.) ± 0.3 |
| XV |  | 42 (h.s.) ± 0.3 | 39 (h.s.) ± 0.3 |

* Note: l.s. – low sensitivity of the culture to the given concentration of the test substance; s. – sensitivity of the culture to the given concentration of the test substance; h.s. – high sensitivity of the culture to the given concentration of the test substance.

ATCC 25923 and *Bacillus subtilis* ATCC 6633 were used. The suspension of the test microorganism was prepared according to the method [10]. Standardization of the bacterial suspension of microorganisms prepared was carried out using a Densi-La-Meter device (manufactured by PLIVA-Lachema, Czech Republic). Synchronization of cultures by changing the cultivation temperature was achieved with a single effect of low temperature (4°C). Microbial load was $10^7$ microbial cells per 1 ml of the medium and set up according to McFarland standard. We worked with 18-24 hour culture of microorganisms.

For studies Mueller-Hinton agar ("HIMedia Laboratories, Pvt. Ltd India" the shelf life of the medium to XI 2016, manufactured by India) was used.

Diffusion of the drug into the agar was conducted by the "wells" method [7]. When assessing the activity

of compounds I-XV, as well as when studying antibiotic-resistant strains the following criteria were used:

- the absence of inhibition zones of microorganisms around the well, the diameter of the inhibition zone to 10 mm indicates that the organism is insensitive to the drug introduced into the well or to the antibiotic concentration;
- inhibition zones with the diameter of 10-15 mm indicate a low sensitivity of the culture to the given concentration of the test substance;
- inhibition zones with the diameter of 15-25 mm assessed as an indicator of the sensitivity of a microorganism to the test substance;
- inhibition zones, which diameter exceeds 25 mm, indicates the high sensitivity of microorganisms to the test substance.

**Results and Discussion**

A low sensitivity of the set of microorganisms used to the action of aliphatic amines I-VI was determined. The antimicrobial activity of aminoalcohols VII-IX was slightly higher, and its enhancement is observed with increasing the number of hydroxyethylene radicals in the molecule. Compounds X-XII containing a carboxyl and hydroxymethyl group in the molecule was more active, with the increasing number of hydroxyethylene groups the inhibition zones were 20-21 mm for *Staphylococcus aureus* and 17-25 mm for *Bacillus subtilis*.

Compounds containing a carboxyl group and methyl radicals appeared to be the most promising among the compounds tested. It is this combination of substituents that contributes to a high microbicide activity in relation to *Staphylococcus aureus* and *Bacillus subtilis*. Increase of the number of methyl radicals in the molecules of compounds XIII-XV leads to a significant increase in activity – the inhibition zones are 39-42 mm.

CONCLUSIONS

1. The effect of various functional groups containing in the molecules of N-, R-alkylamines derivatives on their antibacterial activity in relation to some gram-positive strains of microorganisms has been determined.

2. The aliphatic amines and aminoalcohols studied show a weak or moderate activity in relation to strains of *Staphylococcus aureus* ATCC 25923 and *Bacillus subtilis* ATCC 6633.

3. Compounds containing a carboxyl group and methyl radicals in the molecule exhibit the greatest antimicrobial activity in relation to the gram-positive strains of microorganisms under research.

REFERENCES


1. *Антисептики в профилактике и лечении инфекций / Под ред. Г.К.Палия. – К.: Здоровье, 1997. – С. 108.*
2. *Бобков С.С., Смирнов С.К. Синильная кислота. – М.: Химия, 1970. – С. 172.*
3. *Вейганд – Хильгетаг. Методы эксперимента в органической химии / Пер. с нем. – М.: Химия, 1968. – 944 с.*
4. *Внутрибольничные инфекции: проблемы и решения [Электронный ресурс] / Журнал «Заместитель главного врача»: интернет-журн. – 2008. – Вып. 6. – Режим доступа: http://www.zdrav.ru/articles/practice/detail.php?ID=76435*
5. *Всемирная Организация Здравоохранения 2015. Европейское региональное бюро [Электронный ресурс] – Режим доступа: http://www.euro.who.int/ru/health-topics/disease-prevention/antimicrobial-resistance/antibiotic-resistance/frequently-asked-questions.*
6. *Голодников Г.В., Мандельштам Т.В. Практикум по органическому синтезу. – Л.: Изд-во Ленинградского университета, 1976. – С. 372.*
7. *Державна фармакопея України / Державне підприємство «Науково-експертний фармакопейний центр». – 1-е вид. – Х.: РІРЕГ, 2011. – Доп. 4. – 536 с.*
8. *Практическое руководство по биологической безопасности в лабораторных условиях. 3-е изд. – Всемирная Организация Здравоохранения. Женева, 2004. – 190 с.*
9. *Рабинович В.А., Хавин З.Я. Краткий химический справочник. – 3-е изд., перераб. и доп. – Л.: Химия, 1991. – 432 с.*
10. *Стандартизація приготування мікробних суспензій. Інформаційний лист про нововведення у системі охорони здоров'я України №163-2006. – Київ, 2006.*
11. *Gallant R.W. // Hydrocarbon Processing. – 1973. – Vol. 52, №11. – P. 150.*
12. *Kirk-Othmer encyclopedia, 3-rd ed. – Vol. 1. – N.-Y., 1978. – P. 944.*
13. *Kirk-Othmer encyclopedia, 3-rd ed. – Vol. 2. – N.-Y., 1978. – P. 272-282.*
14. *Wisplinghoff H., Bischoff T., Tallent S.M. et al. // Clin. Infect. Dis. – 2004. – №39. – P. 309-317.*

**ВПЛИВ ЗАМІСНИКІВ В МОЛЕКУЛАХ N-, R-АЛКІЛАМІНІВ НА ДЕЯКІ ГРАМПОЗИТИВНІ ШТАМИ МІКРООРГАНІЗМІВ**

***М.Ю.Голік, М.А.Комісаренко, С.Г.Леонова, Т.П.Осолодченко***

***Ключові слова:*** *N-, R-алкіламіни; антимікробна активність*

*Інтенсивне застосування антибіотиків у лікарських установах, часто без визначення специфіки та чутливості до них, призводить до більш швидкого розвитку стійкості до збудників внутрішньолікарняних інфекцій, що є однією з актуальних проблем охорони здоров'я України. Найбільш розповсюдженими грампозитивними збудниками внутрішньолікарняних інфекцій є Staphylococcus aureus, коагулазонегативні стафілококи та ентерококи. З метою встановлення залежності антимікробної дії від природи замісників в молекулах N-, R-алкіламінів були протестовані деякі метил-, етиламіни, аміноспирти, N-гідроксиметил- та N-метил-N-карбоксиметиламіни. У відповідності до рекомендацій ВООЗ для оцінки антибактеріальної активності N-, R-алкіламінів використовували грампозитивні тест-штами Staphylococcus aureus ATCC 25923 та Bacillus subtilis ATCC 6633. По відношенню до штамів Staphylococcus aureus ATCC 25923 та Bacillus subtilis ATCC 6633 досліджувані аліфатичні аміни та аміноспирти виявляють слабку або помірну активність. Найбільшу антимікробну активність до досліджуваних грампозитивних штамів мікроорганізмів проявляють сполуки, що містять у молекулах карбоксильну групу та метильні радикали.*

**ВЛИЯНИЕ ЗАМЕСТИТЕЛЕЙ В МОЛЕКУЛАХ N-, R-АЛКИЛАМИНОВ НА НЕКОТОРЫЕ ГРАММПОЛОЖИТЕЛЬНЫЕ ШТАММЫ МИКРООРГАНИЗМОВ**

***Н.Ю.Голик, Н.А.Комиссаренко, С.Г.Леонова, Т.П.Осолодченко***

***Ключевые слова:*** *N-, R-алкиламины; антимикробная активность*

*Интенсивное использование антибиотиков в лечебных учреждениях, зачастую без определения специфики и чувствительности к ним, приводит к более быстрому развитию устойчивости к возбудителям внутрибольничных инфекций, что является одной из актуальных проблем здравоохранения Украины. Наиболее распространенными грамположительными возбудителями внутрибольничных инфекций являются Staphylococcus aureus, коагулазонегативные стафилококки и энтерококки. С целью установления зависимости антимикробного действия от природы заместителей в молекулах N-, R-алкиламинов были протестированы некоторые метил-, этиламины, аминоспирты, N-гидроксиметил- и N-метил-N-карбоксиметиламины. В соответствии с рекомендациями ВОЗ для оценки антибактериальной активности N-, R-алкиламинов использовали грамположительные тест-штаммы – Staphylococcus aureus ATCC 25923 и Bacillus subtilis ATCC 6633. По отношению к штаммам Staphylococcus aureus ATCC 25923 и Bacillus subtilis ATCC 6633 исследуемые алифатические амины и аминоспирты проявляют слабую или умеренную активность. Наибольшую антимикробную активность к исследуемым грамположительным штаммам микроорганизмов проявляют соединения, содержащие в молекулах карбоксильную группу и метильные радикалы.*

*Recommended by Doctor of Medicine, professor A.I.Bereznyakova*

UDC 615.275.4:547.822.7:616.36-002:616.127

# THE STUDY OF A POSSIBLE GENERAL TOXICAL ACTION OF 5,7-DIHYDRO-1H-PYRROLO[2,3-d]PYRIMIDINE DERIVATIVES

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*Key words: acute toxicity; chemical substance 5,7-dihydro-1H-pyrrolo[2,3-d]pyrimidine*

*It has been experimentally proven that with the intraperitoneal way of introduction of Dezapur the maximum tolerated dose for mice is 2000 mg/kg, the least toxic dose – 1000 mg/kg, the median lethal dose – 3000 mg/kg. When the dose of 3500 mg/kg was introduced to rats one animal died; in the dose of 5000 mg/kg there was 100% death of animals, the median lethal dose was 4500 mg/kg of the animal's weight. In intragastric introduction the maximum dose of Dezapur for mice was 6000 mg/kg, for rats it was 11000 mg/kg; by the end of the observation period all animals were alive and alert. According to K.K.Sydorov classification the derivative with the code of KMS-211 ("Dezapur") has been proven to belong to practically nontoxic substances (the VI class of toxicity).*

While researching a new drug, along with the study of its medicinal properties, it is mandatory to study the general toxic action to verify the nature of severity of its damaging effects on the body of experimental animals and assess the safety [8].

The study of acute toxicity parameters allows to obtain the necessary information for determining the level of toxicity, the relationship between the dose and adverse effects, and to carry out a comparative assessment of toxicity of new substances with the already known ones, the most effective prototypes or analogues, which are widely used in medical practice. One of the toxicological characteristics of a pharmacological drug is $LD_{50}$ value determined when studying acute toxicity [5, 10].

To study the acute toxicity 15 compounds – derivatives of 5,7-dihydro-1H-pyrrolo[2,3-d]pyrimidine have been selected. They were obtained at the Department of Organic Chemistry by associate professor K.M. Sytnyk under the code of KMS-140, 162, 163, 164, 166, 168, 172, 174, 176, 178, 179, 191, 211, 214, 217. When studying the pharmacological properties (antihypoxic, antioxidant) the compound with the code of KMS-211 conventionally called "Dezapur" has shown the greatest activity [4, 6, 7].

**Materials and Methods**

The median lethal dose was studied in two species of animals: outbred white mice weighing 18-20 g and white rats weighing 180-200 g with two routes of administration – intragastric and intraperitoneal injections. Using two species of animals serves to determine their sensitivity to the drug, which allows us to extrapolate the results to a human [2, 3, 11].

When studying the acute toxicity of "Dezapur" the compound was injected to mice intragastrically in the doses from 3500 mg/kg to 6000 mg/kg of the animal's body weight and intraperitoneally from 1000 mg/kg to 3500 mg/kg; rats were injected intragastric doses from 6000 mg/kg to 11000 mg/kg of the animal's body weight and intraperitoneal doses from 2500 mg/kg to 5000 mg/kg [1, 9].

**Experimental Part**

The results of determination of acute toxicity of "Dezapur" are given in Tab. 1-4.

Table 1

Determination of the acute toxicity of "Dezapur" in mice by intragastric injection (n=6)

| Series | Dose, mg/kg | Surviving animals, number | Mortality of animals | |
|---|---|---|---|---|
| | | | number | % |
| I | 3500 | 6 | 0 | 0.00 |
| II | 4000 | 6 | 0 | 0.00 |
| III | 4500 | 6 | 0 | 0.00 |
| IV | 5000 | 6 | 0 | 0.00 |
| V | 5500 | 6 | 0 | 0.00 |
| VI | 6000 | 6 | 0 | 0.00 |

Table 2

Determination of the acute toxicity of "Dezapur" in rats by intragastric injection (n=6)

| Series | Dose, mg/kg | Surviving animals, number | Mortality of animals | |
|---|---|---|---|---|
| | | | number | % |
| I | 6000 | 6 | 0 | 0.00 |
| II | 7000 | 6 | 0 | 0.00 |
| III | 8000 | 6 | 0 | 0.00 |
| IV | 9000 | 6 | 0 | 0.00 |
| V | 10000 | 6 | 0 | 0.00 |
| VI | 11000 | 6 | 0 | 0.00 |

Table 3

Determination of the acute toxicity of "Dezapur" in mice by intraperitoneal injection (n=6)

| Series | Dose, mg/kg | Surviving animals, number | Mortality of animals | |
|---|---|---|---|---|
| | | | number | % |
| I | 1000 | 6 | 0 | 0.00 |
| II | 1500 | 6 | 0 | 0.00 |
| III | 2000 | 6 | 0 | 0.00 |
| IV | 2500 | 5 | 1 | 16.70 |
| V | 3000 | 3 | 3 | 50 |
| VI | 3500 | 0 | 6 | 100 |

Table 4

Determination of the acute toxicity of "Dezapur" in rats by intraperitoneal injection (n=6)

| Series | Dose, mg/kg | Surviving animals, number | Mortality of animals | |
|---|---|---|---|---|
| | | | number | % |
| I | 2500 | 6 | 0 | 0.00 |
| II | 3000 | 6 | 0 | 0.00 |
| III | 3500 | 5 | 1 | 16.70 |
| IV | 4000 | 4 | 2 | 33.33 |
| V | 4500 | 3 | 3 | 50 |
| VI | 5000 | 0 | 6 | 100 |

The data presented in Tab. 1-2 indicate that with the intragastric administration the maximum possible doses of "Dezapur" are 6000 mg/kg in mice and 11000 mg/kg in rats. By the end of the observation period all animals remained alert, active, with good appetite and shiny hair.

Analysis of the results (Tab. 3-4) indicates that the most tolerated dose for mice is 2000 mg/kg, the least toxic dose is 1000 mg/kg, the median lethal dose is 3000 mg/kg.

With intraperitoneal administration of "Dezapur" to rats in the dose of 3500 mg/kg one animal died; in the dose of 5000 mg/kg there was 100% death. The median lethal dose is 4500 mg/kg of the animal's body weight.

CONCLUSIONS

According to K.K. Sydorov classification and taking the route of administration into account "Dezapur" refers to practically nontoxic substances (the VI class of toxicity).

REFERENCES


1. *Доклінічні дослідження лікарських засобів: Метод. рекоменд. / Під ред. О.В.Стефанова. – К.: Авіценна, 2001. – 528 с.*
2. *Использование лабораторных животных в токсикологическом эксперименте: Метод. рекоменд. / Под ред. проф., акад. РАМН П.И.Сидорова. – Архангельск, 2002. – 15 с.*
3. *Штабский Б.М., Красовский Г.Н., Кудрина В.Н. и др. // Гигиена и санитария. – 1980. – №10. – С. 41-44.*
4. *Caro A.A., Cererbum A.I. // Annu Rev. Pharmacol. Toxicol. – 2004. – Vol. 44. – P. 27-42.*
5. *Giri D.K., Ghosh R.C., Mondal M. // Ind. J. of Veterinary Pathol. – 2014. – Vol. 38 (1). – P. 33-38.*
6. *Maes M., Fisar Z., Medina M., Scapagnini G. // Inflammopharmacol. – 2012. – Vol. 20(3). – P. 127-150.*
7. *Matalka I.I., Fatlauri L.A., Mhaidat N.M. // Intern. J. of Physiol., Pathophysiol. and Pharmacol. – 2013. – Vol. 5, №2. – P. 102-108.*
8. *Morales G., Peredes A., Olivares A. et al. // Biol. Res. – 2014. – Vol. 47 (6). – P. 10-17.*
9. *Porter S.N., Howard G.S., Butler R.N. // Eur. J. of Pharmacol. – 2000. – №397. – P. 1-9.*
10. *Tomohiro I., Tomoko O. // Regulatory Toxicol. and Pharmacol. – 2014. – Vol. 68 (2). – P. 240-249.*
11. *Weiss R.F., Fintelmann V. Herbal Medicine. – Stuttgart – New York: Thieme, 2000. – 438 p.*


**ВИЗНАЧЕННЯ МОЖЛИВОЇ ЗАГАЛЬНОТОКСИЧНОЇ ДІЇ ПОХІДНИХ 5,7-ДИГІДРО-1Н-ПІРОЛО[2,3-d]ПІРИМІДИНУ**

***О.В.Севрюков, В.А.Волковой, С.В.Колісник, К.М.Ситнік***

***Ключові слова:*** *гостра токсичність; похідні 5,7-дигідро-1Н-піроло[2,3-d]піримідину*

*Гостру токсичність похідних вивчали на лабораторних тваринах (мишах, щурах) при двох шляхах введення (внутрішньошлунковому, внутрішньоочеревинному). Експериментально доведено, що при внутрішньоочеревинному введенні Дезапуру найкраще переносима доза для мишей становить 2000 мг/кг, найменш токсична доза – 1000 мг/кг, середньосмертельна доза – 3000 мг/кг. При введенні щурам у дозі 3500 мг/кг загинула 1 тварина, а у дозі 5000 мг/кг загинуло 100% тварин, середньосмертельна доза складала 4500 мг/кг маси тварини. При внутрішньошлунковому введенні максимально можливої дози Дезапуру мишам 6000 мг/кг, щурам 11000 мг/кг усі тварини до кінця терміну спостереження залишалися живими і бадьорими. Встановлено, що похідне під шифром KMS-211 (Дезапур) за класифікацією К.К. Сидорова з урахуванням шляху введення належить до практично нетоксичних речовин (VI клас токсичності).*

**ИЗУЧЕНИЕ ВОЗМОЖНОГО ОБЩЕТОКСИЧЕСКОГО ДЕЙСТВИЯ ПРОИЗВОДНЫХ 5,7-ДИГИДРО-1Н-ПИРРОЛО [2,3-d]ПИРИМИДИНА**

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***Ключевые слова:*** *острая токсичность; производные 5,7-дигидро-1Н-пирроло[2,3-d]пиримидина*

*Острую токсичность производных изучали на лабораторных животных (мышах, крысах) при двух путях введения (внутрижелудочном, внутибрюшинном). Экспериментально доказано, что при внутрибрюшинном введении Дезапура наиболее переносимая доза для мышей составляет 2000 мг/кг, наименее токсичная доза – 1000 мг/кг, среднесмертельная доза – 3000 мг/кг. При введении крысам в дозе 3500 мг/кг погибло одно животное, а в дозе 5000 мг/кг наблюдалось 100% гибели животных, среднесмертельная доза составляла 4500 мг/кг массы животного. При внутрижелудочном введении максимально возможной дозы Дезапура мышам 6000 мг/кг, крысам 11000 мг/кг все животные до конца срока наблюдения оставались живыми и бодрыми. Доказано, что производное под шифром KMS-211 (Дезапур) по классификации К.К.Сидорова с учетом пути введения относится к практически нетоксичным веществам (VI класс токсичности).*

*Recommended by Doctor of Medicine, professor S.Yu.Shtrygol'*

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# THE HYPOLIPIDEMIC ACTIVITY OF *POTERIUM SANGUISORBA L.*

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*Key words: hypolipidemic drugs; plant extracts; experiments on animals*

*Atherosclerosis is recognized as "non-infectious pandemic of the XXI-th century". There is a wide range of drugs for correction of hyperlipidemia, as well as for the primary and secondary prevention of atherosclerosis complications. However, all known lipid-lowering drugs have many side effects. Herbal medicines have several advantages over synthetic monodrugs. In particular, due to the complex and balanced chemical composition and the rational combination of biologically active substances they have multifaceted effects on the body: on the one hand, they affect directly on the diseased area, and on the other hand, provide pharmacological correction of various functional systems. In this regard it is interesting to study triterpenoid compounds isolated from the plant material, which have a wide range of pharmacological activities, including the lipid-lowering effect. The aim of this study was to assess the lipid-lowering properties of the ethanol extract from Poterium sanguisorba L. (burnet salad) containing triterpenoid compounds in the underground part of the plant. The ethanol extract of Poterium sanguisorba L. underground part was studied for the hypolipidemic activity in the acute model of hyperlipidemia induced by injecting a single dose of Tween-80 (250 mg/kg) intraperitonially to rats. In 8 h the blood samples were taken, and the level of hyperlipidemia was assessed. Daily white mature Wistar rats were orally treated with the extract in two doses: 100 mg/kg and 250 mg/kg for 7 days, the last injection was together with Tween-80. As a reference drug nicotinic acid (30 mg/kg) was used. At the end of the study the serum levels of the total cholesterol, triglycerides and the total lipids were measured. Determination was performed using enzymatic methods. The results of the study conducted have confirmed the hypolipidemic activity of Poterium sanguisorba L. extract under conditions of acute hyperlipidemia.*

Atherosclerosis is the traditionally known consequence of the lipid metabolism disorders (dyslipidemia, hyperlipidemia, hypertriglyceridemia) and reduction of the elasticity of vessels of different caliber [4]. Hyperlipidemia is the major risk factor for atherosclerosis. Other complications are coronary heart disease, ischemic cerebrovascular disease, hypertension, obesity and diabetes mellitus (Type II). Although many effective lipid-lowering synthetic drugs exist, none is effective for all lipoprotein disorders, and all of them are associated with some adverse effects. Therefore, it is a need of the day to search other products from natural sources that are less toxic, less expensive, and can provide better safety and efficacy when using. Natural products from plants are a rich source used for centuries to cure various ailments [12].

Today, a lipid-lowering therapy uses a wide range of drugs (statins, fibrates, cholesterol absorption inhibitors, niacin drugs) for correction of hyperlipidemia, as well as the primary and secondary prevention of atherosclerosis complications [5]. The known lipid-lowering drugs have many side effects in patients [11].

Herbal medicines have several advantages over synthetic monodrugs. In particular, due to the complex and balanced chemical composition and the rational combination of biologically active substances they have multifaceted effects on the body: on the one hand, they affect directly on the diseased area, and on the other hand, provide pharmacological correction of various functional systems [3, 13]. In this regard it is interesting to study triterpenoid compounds isolated from the plant material, which have a wide range of pharmacological activities, including the lipid-lowering effect [1, 10]. The *Rosaceae* family includes such interesting species as *Poterium polygamum Waldst. Et Kit*., *P. sanguisorba L., P. lasiocarpum Boiss. Et Hausskn.*, which are widespread in Ukraine. Such triterpenoids as caccigenin, poteriside and tormentoside are contained in the underground part of these plants [6].

The aim of this study was to assess the antihyperlipidemic properties of the ethanol extract of *P. sanguisorba L*. (burnet salad) on the model of acute hyperlipidemia.

**Materials and Methods**

Only male albino Wistar rats weighing 200-220 g were used in this study. They were acclimatized under laboratory conditions. Animals were fed on a standard diet and water *ad libitum*. Thirty animals were distributed into 5 groups (six rats in each group): Group 1 – control rats, group 2 – intact rats, group 3 – standard rats, group 4-5 – test rats.

*P. sanguisorba L*. rhizomes were collected in the Zaporizhzhia region, dried under shade, powdered and extracted with 96% ethanol in a Soxhlet extractor. The extract was concentrated using a vacuum evaporator, and the extract residue was stored at 4°C in refrigerator for further use.

Table

The effect of *Poterium sanguisorba L.* extract on plasma lipids on the acute model of hyperlipidemia

| Group | Total cholesterol (mmol/l) | Triglycerides (mmol/l) | Total lipids (g/l) |
|---|---|---|---|
| Intact rats | 1.19±0.002* | 0.26±0.004** | 6.56±0.209** |
| *% to control rats* | ***86.86%*** | ***66.66%*** | ***73.29%*** |
| Control group | 1.37±0.064 | 0.39±0.02 | 8.95±0.234 |
| PSE 100 mg/kg | 1.12±0.075* | 0.28±0.003** | 5.2±0.624** |
| *% to control rats* | ***81.75%*** | ***71.79%*** | ***58.10%*** |
| PSE 250 mg/kg | 1.26±0.144 | 0.31±0.009** | 5.35±0.25** |
| *% to control rats* | ***91.97%*** | ***79.48%*** | ***59.77%*** |
| Nicotinic acid | 1.25±0.12 | 0.30±0.02* | 5.02±0.471** |
| *% to control rats* | ***91.24%*** | ***76.92%*** | ***56.09%*** |

Notes: * – $p<0.05$; ** – $p<0.01$ – probability differences compared to the control group.

The model of acute hyperlipidemia is based on the ability of such surfactant as Tween-80 to bind blood plasma lipids and to form micelles isolated from the action of lipoprotein lipase. In 8-10 h after a single intraperitoneal administration of Tween-80 the level of blood triglycerides increased by 6-8 times and the level of cholesterol increased by 3-4 times.

The model of acute hyperlipidemia was induced by injecting a single dose of Tween-80 intraperitonially in rats in the amount of 250 mg/kg per 1 ml of distilled water [8]. In eight hours the blood samples were collected to assess the level of hyperglycemia. Blood was collected by decapitation of anesthetized rats (sodium thiopental, 40 mg/kg) and centrifuged at 3000 rpm for 15-20 min. The serum samples were further subjected to biochemical analysis.

In acute toxicity studies [7] it was determined that a single oral (by gavage) introduction of *P. sanguisorba L*. extract (PSE) in the dose of 5 g/kg did not cause the death of animals and did not also contribute to the negative impact on their condition and behaviour. Thus, PSE can be referred to the IV-th class of toxicity (low-toxic compounds). In this study PSE was administered in two doses of 100 and 250 mg/kg (Group 4-5, respectively) introduced in advance within 7 days before the injection of Tween-80.

The antihyperlipidemic activity of PSE was compared to nicotinic acid (30 mg/kg) [9] administered intraperitonially to standard rats (Group 3).

At the end of the study the total cholesterol (TC), triglycerides (TG) and the total lipids (TL) were measured in the serum. Determination was performed using enzymatic methods (Felicit, Ukraine).

Statistical analysis was performed using Student t-test. The data obtained was processed by the method of variation statistics at the significance level of $p<0.05$.

**Results and Discussion**

The data in Table show that a single administration of Tween-80 caused marked increase in levels of TC (13.14%, $p<0.05$), TG (33.34%, $p<0.05$) and TL (26.71%, $p<0.05$) in the plasma in control rats compared to intact rats.

Treatment with PSE caused decrease in these levels in both doses. But the dose of 100 mg/kg showed the highest reduction.

Thus, PSE in the dose of 100 mg/kg reduced the level of TC by 18.25% compared to the control group, while PSE in the dose of 250 mg/kg and nicotinic acid reduced the probability of the appropriate level by 8.03% and 8.76%, respectively.

Besides, administration of PSE in the dose of 100 mg/kg caused the highest TG level decrease in the blood (28.21%) and appeared to be 5.13% higher than in standard rats receiving nicotinic acid. Increase in the dose of PSE to 250 mg/kg did not result in increase of its effect (reduction of the blood TG level compared to control is 20.52%).

Under the effect of PSE the level of TL significantly decreased compared to the control to almost identical values. Thus, in Group 4 the level of TL decreased by 41.90% and in Group 5 by 40.23%. Nicotinic acid showed the more pronounced effect on overall blood lipids, thus reducing the corresponding figure by 43.91%.

Therefore, we have found that the ethanol extract of *Poterium sanguisorba L*. on the model of acute hyperlipidemia showed a pronounced hypolipidemic activity. The ability to reduce the blood lipid spectrum – the total cholesterol, triglycerides, and the total lipids – was manifested. However, a significant increase in the efficiency of PSE when increasing the dose was not found. Therefore, the dose of 100 mg/kg was determined to be the most effective lipid-lowering dose in this study. The ability of PSE to reduce the plasma levels of TC, TG and TL is close to nicotinic acid.

CONCLUSIONS

1. The results of the study show that *P. sanguisorba L*. has a significant antihyperlipidemic action against experimentally-induced hyperlipidemia.

2. It has been found that the most effective dose in the conditions of acute hyperlipidemia is the dose of 100 mg/kg.

3. The lipid profile under action of PSE (100 mg/kg) can be comparable with nicotinic acid.

REFERENCES


1. *Горбатюк Н.О., Назарова Л.Е., Сергеева Е.О., Саджая Л.А. // Мед. вестник Башкортостана. – 2011. – №4. – С. 108-110.*
2. *Западнюк И.П., Западнюк В.И., Захария Е.А., Западнюк Б.В. Лабораторные животные. Разведение, содержание, использование в эксперименте. – К.: Вища школа, 1983. – 383 с.*
3. *Мухаммед А.А., Максимов М.Л. // Вестник новых мед. технол. – 2014. – №1. – С. 1-14.*
4. *Недогода С.В., Морозова Т.Е. // Артериальная гипертензия. – 2012. – №1. – С. 5-10.*
5. *Ратькин А.В., Кайдаш О.А., Иванов В.В. и др. // Бюл. сибирской медицины. – 2014. – №1. – С. 67-72.*
6. *Соколов П.Д., Барнаулов О.Д., Кузнецова Г.А., Медведева Л.И. Растительные ресурсы СССР: Цветковые растения, их химический состав, использование; Семейства Hydrangeaceae – Haloragaceae. – Л.: Наука, 1987. – 326 с.*
7. *Стефанов А.В. Доклинические исследования лекарственных средств: Метод. рекоменд. / Під ред. А.В.Стефанова. – К., 2002. – С. 84-90.*
8. *Сусеков А.В., Зубарева М.Ю., Кухарчук В.В. // Клин. фармакол. и терапия. – 2002. – №1. – С. 71-74.*
9. *Хабриев Р.У. Руководство по экспериментальному (доклиническому) изучению новых фармакологических веществ. – М.: Медицина, 2005. – 832 с.*
10. *Bailey C., Day C. // Diabetes Care. – 1989. – Vol. 12, №3. – P. 553-564.*
11. *Kumar Vishnu, Singh Pradyumn, Chander Ramesh // Ind. J. of Biochem. & Biophysics. – 2009. – Vol. 46. – P. 507-510.*
12. *Srinivasa Rao Brahma // Intern. J. of Res. in Pharmacy and Chemistry. – 2013. – Vol. 3 (3). – P. 708-711.*
13. *Upendra Rao M. // Intern. J. of Chem. Tech. Res. – 2010. – №2 (3). – P. 1883-1892.*


**ГІПОЛІПІДЕМІЧНА АКТИВНІСТЬ ЧОРНОГОЛОВНИКА РОДОВИКОВОГО**

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***Ключові слова:*** *гіполіпідемічні засоби; екстракти з рослин; експерименти на тваринах*

*Атеросклероз визнаний «неінфекційною пандемією XXI століття». Існує широкий спектр лікарських засобів для корекції гіперліпідемії, а також для первинної та вторинної профілактики ускладнень атеросклерозу. Проте усі відомі гіполіпідемічні препарати мають безліч побічних ефектів. Рослинні препарати мають ряд переваг перед синтетичними монопрепаратами. Завдяки складному та збалансованому хімічному складу, раціональному поєднанню біологічно активних речовин вони чинять багатосторонню дію на організм: впливають, з одного боку, безпосередньо на вогнище ураження, а з іншого боку, забезпечують фармакологічну корекцію різних функціональних систем. Становить інтерес вивчення тритерпеноїдних сполук, виділених з рослинної сировини, які мають широкий спектр фармакологічної активності, в тому числі гіполіпідемічну дію. Мета даного дослідження полягала в оцінці гіполіпідемічних властивостей етанольного екстракту з чорноголовника родовикового (P. sanguisorba L)., який містить у підземній частині тритерпеноїдні сполуки. Білим статевозрілим щурам лінії Вістар однократно внутрішньоочеревинно вводили твін-80 з розрахунку 250 мг/кг в 1 мл води дистильованої. Рівень гіперліпідемії оцінювали через 8 годин після введення твіну-80, взявши зразки крові. Перорально вводили екстракт у 2-х дозах: 100 мг/кг та 250 мг/кг заздалегідь протягом 7 днів, проводячи останнє введення разом з твіном-80. Як препарат порівняння використали нікотинову кислоту (30 мг/кг). В кінці дослідження у сироватці були виміряні рівні загального холестерину, тригліцеридів та загальних ліпідів. Визначення проводили з використанням ферментативних методів. Результати проведеного дослідження підтвердили наявність гіполіпідемічної активності екстракту підземних органів чорноголовника родовикового в умовах індукованої гострої гіперліпідемії.*

**ГИПОЛИПИДЕМИЧЕСКАЯ АКТИВНОСТЬ ЧЕРНОГОЛОВНИКА КРОВОХЛЕБКОВОГО**

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***Ключевые слова:*** *гиполипидемические средства; экстракты из растений; эксперименты на животных*

*Атеросклероз признан «неинфекционной пандемией XXI века». Существует широкий спектр лекарственных средств для коррекции гиперлипидемии, а также для первичной и вторичной профилактики осложнений атеросклероза. Однако все известные гиполипидемические препараты имеют множество побочных эффектов. Растительные препараты имеют ряд преимуществ перед синтетическими монопрепаратами. Благодаря сложному и сбалансированному химическому составу, рациональному сочетанию биологически активных веществ они оказывают многостороннее действие на организм: влияют, с одной стороны, непосредственно на очаг поражения, а с другой стороны, обеспечивают фармакологическую*

*коррекцию различных функциональных систем. Представляет интерес изучение тритерпеноидных соединений, выделенных из растительного сырья, которые обладают широким спектром фармакологической активности, в том числе гиполипидемическим действием. Цель данного исследования заключалась в оценке гиполипидемических свойств этанольного экстракта черноголовника кровохлебкового (P. sanguisorba L)., который содержит в подземной части тритерпеноидные соединения. Белым половозрелым крысам линии Вистар однократно внутрибрюшинно вводили твин-80 из расчета 250 мг/кг в 1 мл воды дистиллированной. Уровень гиперлипидемии оценивали через 8 часов после введения твина-80, взяв образцы крови. Перорально вводили экстракт в 2-х дозах 100 мг/кг и 250 мг/кг заранее в течение 7 дней, проводя последнее введение вместе с твин-80. В качестве препарата сравнения использовали никотиновую кислоту (30 мг/кг). В конце исследования в сыворотке были измерены уровни общего холестерина, триглицеридов и общих липидов. Определение проводили с использованием ферментативных методов. Результаты проведенного исследования подтвердили наличие гиполимидемической активности экстракта подземных органов черноголовника кровохлебкового в условиях индуцированной острой гиперлипидемии.*

# ПРАВИЛА ПІДГОТОВКИ МАТЕРІАЛІВ ДО ПУБЛІКАЦІЇ В ЖУРНАЛІ "ВІСНИК ФАРМАЦІЇ"

*Загальні положення*

Журнал «Вісник фармації» публікує оригінальні статті, присвячені теоретичним та практичним досягненням у галузі фармації.

До розгляду приймаються оригінальні статті (до 10 сторінок), присвячені проблемам управління та економіки фармації, синтезу, аналізу, технології, дослідженню біологічної активності фізіологічно активних речовин та лікарських препаратів, експериментальній та клінічній фармакології, що містять теоретичні або експериментальні результати досліджень, які не були опубліковані раніше.

Статті, відіслані авторам на виправлення, повинні бути повернені до редакції не пізніше, ніж через 2 тижні після одержання. При перевищенні зазначеного строку рукопис буде перереєстрований як такий, що надійшов знову з відповідною зміною дати його виходу у світ. В авторській коректурі допускається виправлення лише помилок набору.

Статті повинні містити такі елементи: постановка проблеми у загальному вигляді та її зв'язок із важливими науковими чи практичними завданнями; аналіз останніх досліджень і публікацій, в яких започатковано розв'язання даної проблеми і на які спирається автор, виділення невирішених раніше частин загальної проблеми, яким присвячується означена стаття; формулювання цілей статті (постановка завдання); виклад основного матеріалу дослідження з повним обґрунтуванням отриманих наукових результатів; висновки з даного дослідження і перспективи подальших розробок у даному напрямку.

*Представлення статей*

Статті подаються до редакції у двох екземплярах і супроводжуються направленням від організації, в якій виконана робота, на ім'я головного редактора та експертним висновком, який дозволяє відкриту публікацію. Другий екземпляр статті підписується всіма авторами.

Автори статей, поданих до редакції для публікації в журналі, своїми особистими підписами на примірниках рукописів статей засвідчують:

- згоду на ведення редакцією обліку необхідних для обробки статей особистих даних авторів (ПІБ, учене звання, учений ступінь, посада та місце роботи, адреса для листування, робочий телефон, електронна пошта) з метою забезпечення відносин у сфері права інтелектуальної власності, в тому числі авторського права;
- дозвіл на публікацію особистих даних авторів (ПІБ, учене звання, учений ступінь, місце роботи, робочий телефон, електронна пошта) в журналі разом зі статтею;
- згоду на оприлюднення повної електронної версії статті (або рефератів статті) на сайтах Національного фармацевтичного університету, Національної бібліотеки України ім. В.І.Вернадського та інших порталах наукової періодики з обов'язковим зазначенням та збереженням особистих немайнових авторських прав.

До рукопису додається дискета у форматі MS Word, яка містить ідентичний матеріал.

До статті на окремому аркуші додаються відомості про авторів, які містять: учене звання, учений ступінь; прізвище, ім'я та по батькові (повністю); місце роботи та посаду, яку обіймає автор; адресу, номери телефонів і факсів, E-mail для листування.

Додатково може бути вислана електронна версія рукопису (за адресою: press@ukrfa.kharkov.ua)

*Оформлення рукописів*

Текст статті друкується кеглем №14 через 1,5 інтервали на аркуші формату А4 (ширина полів: зліва – 3 см, справа – 1 см, зверху та знизу – по 2 см) і починається з таких даних: УДК, назви статті, ініціалів та прізвищ всіх авторів, назви організацій, в яких виконана робота, переліку ключових слів (понять) у кількості 5-8 українською, російською, англійською мовами.

Автори повинні дотримуватись загального плану побудови статті.

1. Вступ. Містить постановку проблеми, короткий огляд раніше надрукованих робіт у досліджуваній галузі, зазначається актуальність тематики, мета роботи.

2. Експериментальна частина (Матеріали та методи). Містить описання використаних або розроблених методик, приладів та умов вимірювання. В хімічних методиках вказують кількість реагентів у мольних та масових одиницях (для каталізаторів – масу та мольні відсотки), об'єми розчинників, кількість та виходи одержаних сполук. Для всіх вперше синтезованих сполук повинні бути наведені дані елементного аналізу або мас-спектра високого розрізнення. В емпіричних брутто-формулах елементи розміщують за системою Chemical Abstracts: C, H та далі згідно з латинським алфавітом.

3. Результати та їх обговорення. Містять результати досліджень, зроблених автором. Зміст роботи необхідно викладати ясно та стисло, уникаючи відомих положень, повторення результатів у тексті, таблицях і рисунках. Для хімічних сполук, вперше описаних у статті, або тих, що є основним об'єктом дослідження, крім формули наводиться повна назва згідно з номенклатурою IUPAC.

4. Висновки.

5. Перелік використаної літератури, розташованої за алфавітом (спочатку кирилиця, потім – латинський шрифт). Пристатейний список літератури повинен містити публікації за останні 10 років. Більш ранні публікації допускаються лише в особливих випадках; 60% літературних джерел повинні бути іноземною мовою.

На кожну роботу у списку літератури повинна бути зроблена відсилка в тексті рукопису.

*Приклади оформлення літературних джерел:*

1. Логачев В.К. // Вісник фармації. – 2002. – №2 (30). – С. 50-51.

2. Garg R., Kurup A., Hansch C. // Crit. Rev. Toxicol. – 2001. – Vol. 31, №2. – P. 223-245.

3. Erdlenbruch B., Pekrun A., Schiffmann H. et al. // Med. Pediatr. Oncol. – 2002. – Vol. 38, №5. – P. 349-352.

4. Заморський І.І., Кметь О.Г. // Тези доп. наук. конф. «Вікові аспекти чутливості організму до ксенобіотиків». – Чернівці: Медик, 2002. – С. 6.

5. Палій Г.К. Антисептики у профілактиці і лікуванні інфекцій. – К.: Здоров'я, 1997. – 193 с.

6. Андронати С.А. Гидазепам. – К.: Наукова думка, 1992. – 200 с.

7. Лапач С.Н., Чубенко А.В., Бабич П.Н. Статистические методы в медико-биологических исследованиях с использованием Excel. – К.: МОРИОН, 2000. – 320 с.

8. Витворт Дж.А., Лоренс Дж.Р. Руководство по нефрологии / Пер. с англ. – М.: Медицина, 2000. – 485 с.

9. Терапевтический справочник Вашингтонского университета / Пер. с англ. под ред. М.Вудли, А.Уэлана. – М.: Практика, 1995. – 832 с.

10. Тихонов А.И. Разработка технологии и исследование лекарственных форм с фенольными соединениями прополиса: Автореф. дис. ... д-ра фарм. наук. – Х., 1983. – 45 с.

11. Коваленко С.М. Синтез, будова та властивості дво- і триланкових ансамблів циклів з термінальними кумариновими ланками: Дис. ... д-ра хім. наук. – Х., 1993. – 452 с.

12. А.с. 1489778 СССР, МКИ[3] А 61 К 31/425 // Открытия. Изобретения. – 1989. – №24.

13. Пат. 5925616 США, МКИ[6] А 61 К 38/00, А 61 К 31/535. – Опубл.: 20.07.99; НКИ 514/2.

14. Гайдукевич А.Н., Свечникова Е.Н., Костина Т.А. // Деп. в УкрНИИНТИ 19.02.90. №266-Ук90 (Харьк. гос. фарм. ин-т). – Х., 1990. – 4 с.

Опис електронних ресурсів здійснюють згідно з правилами, представленими у бюлетені ВАК України.

Представлені приклади наведені у виданні «Бюлетень ВАК України». – 2009. – №5. – С. 30.

1. Богомольний Б.Р. Медицина екстремальних ситуацій [Електронний ресурс] : навч. посіб. для студ. мед. вузів III-IV рівнів акредитації / Б.Р.Богомольний, В.В.Кононенко, П.М.Чуєв. – 80 Min / 700 MB. – Одеса: Одес. мед. ун-т, 2003. – (Бібліотека студента-медика) – 1 електрон. опт. диск (CD-ROM); 12 см. – Систем. вимоги: Pentium; 32 Mb RAM; Windows 95, 98, 2000, XP; MS Word 97-2000. – Назва з контейнера.

2. Розподіл населення найбільш численних національностей за статтю та віком, шлюбним станом, мовними ознаками та рівнем освіти [Електронний ресурс]: за даними Всеукр. перепису населення 2001 р. / Держ. ком. статистики України; ред. О.Г.Осауленко. – К.: CD-вид-во «Інфодиск», 2004. – 1 електрон. опт. диск (CD-ROM): кольор.; 12 см. – (Всеукр. перепис населення, 2001). – Систем. вимоги: Pentium-266; 32 Mb RAM; CD-ROM Windows 98/2000/NT/XP. – Назва з титул. екрану.

3. Бібліотека і доступність інформації у сучасному світі: електронні ресурси в науці, культурі та освіті: (підсумки 10-ї Міжнар. конф. «Крим-2003») [Електронний ресурс] / Л.Й.Костенко, А.О.Чекмарьов, А.Г.Бровків, І.А.Павлуша // Бібліотечний вісник. – 2003. – №4. – С. 43. – Режим доступу до журн.: http://www.nbuv.gov.ua/articles/2003/03klinko.htm.

Стаття супроводжується трьома рефератами українською, російською та англійською мовами у вигляді розширеної анотації обсягом 200-220 слів. Реферати повинні містити індекс УДК, назву статті, ініціали та прізвища всіх авторів, назву установ (-и). Реферати мають бути інформативними (не містити лише загальні фрази), змістовними, структурованими (повторювати логіку описання результатів у статті), лаконічними і чіткими, з переконливими формулюваннями.

Формули сполук подаються окремими файлами у форматі Corel Draw 13; Chem Win, ISISdraw; діаграми та рисунки – у форматі Excel або Corel Draw 13; рисунки у вигляді фотографій можуть бути представлені файлами TIFF 300-600dpi Gray Scale (256 градацій сірого). Ширина графічного матеріалу повинна бути розміром 8,4 см або 17,4 см. Зображення на рисунках та в таблицях структурних формул небажане.

У статтях повинна використовуватись система одиниць СІ.

Рисунки та підписи до них виконують окремо один від одного; підписи до всіх рисунків статті подаються на окремому аркуші. На зворотному боці кожного рисунка простим олівцем вказується його номер та назва статті, а в разі необхідності – верх і низ.

Таблиці повинні бути надруковані на окремих аркушах і мати нумерацію і заголовки. На полях рукопису необхідно вказати місце розміщення рисунків і таблиць. Інформація, наведена у таблицях і на рисунках, не повинна дублюватися.

Рукописи, оформлені без дотримання вказаних правил, редакція не реєструє та не повертає авторам.

Передплатний індекс у каталозі «Укрпошта» для індивідуальних передплатників 74102, для підприємств – 74103.

# ЗМІСТ / CONTENTS / СОДЕРЖАНИЕ

## СИНТЕЗ ТА АНАЛІЗ БІОЛОГІЧНО АКТИВНИХ РЕЧОВИН



## ТЕХНОЛОГІЯ ЛІКАРСЬКИХ ПРЕПАРАТІВ



## ОРГАНІЗАЦІЯ ТА ЕКОНОМІКА ФАРМАЦІЇ

## ЕКСПЕРИМЕНТАЛЬНА ФАРМАКОЛОГІЯ



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