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THE RESERVED-PHASE HPLC STUDY OF THE COMPLEXATION OF 5,17-BIS-(N-TOLYLIMINO-METHYL)-25,27-DIPROPOXYCALIX[4]ARENE WITH AROMATIC CARBOXYLIC ACIDS

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Key words: Calix[4]arene; reversed-phase high performance liquid chromatography; aromatic carboxylic acids; molecular modelling; Host-Guest complexation

The Host-Guest complexation of 5,17-bis-(N-tolyliminomethyl)-25,27-dipropoxycalix[4]arene with a number of aromatic carboxylic acids has been studied by reversed-phase high-performance liquid chromatography. The mobile phase was acetonitrile-water (80/20, v/v) with addition of 0.1% formic acid. The column was LiChrosorb RP 18, the UV detector operated at λ = 254 nm and at 26°C. The main chromatographic characteristics (retention time t_R and capacity factor k') of the aromatic carboxylic acids have been determined. The lipophilicity values of log P of carboxylic acids, as well as the binding constants K_A (387-941 M^{-1}) and Gibbs free energies ∆G (-14.74 – -16.94 kJ/mol) of the calixarene complexes with aromatic carboxylic acids have been calculated. The molecular modelling (Hyper Chem, version 8.0) of the calixarene complexes has revealed the presence of hydrogen bonds between carboxylic groups of the acids and nitrogen atoms of imino groups at the upper rim or oxygen atoms of hydroxyl groups at the lower rim of the calixarene macrocycle. The influence of log P lipophilicity of acids on K_A values of the calixarene complexes has been assessed. The linear dependence of the binding constants on the acid lipophilicity indicates a significant role of solvophobic interactions on the complexation process. The relationship between supramolecular (K_A) and physicochemical (molecular weight, log P, pKa) characteristics of acids has been found. The binding constants K_A of the complexes increase with increase of their molecular weight and log P values.

ОБЕРНЕНО-ФАЗНЕ ВЕРХ-ДОСЛІДЖЕННЯ КОМПЛЕКСОУТВОРЕННЯ 5,17-БІС-(N-ТОЛІЛІМІНОМЕТИЛ)-25,27-ДИПРОПОКСИКАЛІКС[4]АРЕНУ З АРОМАТИЧНИМИ КАРБОНОВИМИ КИСЛОТАМИ
О.І.Кальченко, А.В.Соловйов, В.І.Кальченко
Ключові слова: *калікс[4]арен; обернено-фазна високоефективна рідинна хроматографія; ароматичні карбонові кислоти; молекулярне моделювання; комплексоутворення типу Гість-Господар*
Комплексоутворення типу Гість-Господар 5,17-біс-(N-толілімінометил)-25,27-дипропоксикалікс[4]арену з низкою ароматичних карбонових кислот досліджено методом обернено-фазної високоефективної рідинної хроматографії. Рухома фаза – ацетонітрил-вода, (80/20, за об'ємом) з додаванням 0,1% мурашиної кислоти. Колонка – LiChrosorb RP 18, УФ детектор працював при довжині хвилі λ = 254 нм за температури 26°С. Визначені основні хроматографічні характеристики (час утримання t_R та коефіцієнт ємкості k') карбонових кислот. Розраховані значення ліпофільності log P кислот, а також значення констант зв'язування K_A (387-941 M^{-1}) та вільних енергій Гібса ∆G (-14.74 – -16.94 kJ/mol) комплексів каліксарену з ароматичними карбоновими кислотами. Молекулярне моделювання (Hyper Chem, версія 8.0) каліксаренових комплексів вказало на присутність водневих зв'язків між карбоксильними групами кислот та атомами азоту іміно-груп на верхньому вінці або атомами кисню гідроксильних груп на нижньому вінці каліксаренового макроциклу. Оцінено вплив ліпофільності log P кислот на значення K_A каліксаренових комплексів. Лінійна залежність констант зв'язування K_A від ліпофільності кислот вказує на суттєву роль сольвофобних взаємодій у процесі комплексоутворення. Встановлено взаємозв'язок між супрамолекулярними (K_A) та фізико-хімічними характеристиками (молекулярна маса, log P) кислот. Константи зв'язування K_A комплексів зростають зі збільшенням молекулярної маси кислот та їх ліпофільності log P.

ОБРАЩЕННО-ФАЗНОЕ ВЭЖХ ИССЛЕДОВАНИЕ КОМПЛЕКСООБРАЗОВАНИЯ 5,17-БИС-(N-ТОЛИЛИМИНОМЕТИЛ)-25,27-ДИПРОПОКСИКАЛИКС[4]АРЕНА С АРОМАТИЧЕСКИМИ КАРБОНОВЫМИ КИСЛОТАМИ
О.И.Кальченко, А.В.Соловйов, В.И.Кальченко
Ключевые слова: *каликс[4]арен; обращенно-фазная высокоэффективная жидкостная хроматография; ароматические карбоновые кислоты; молекулярное моделирование; комплексообразование типа Гость-Хозяин*
Комплексообразование типа Гость-Хозяин 5,17-бис-(N-толилиминометил)-25,27-дипропоксикаликс[4]арена с некоторыми ароматическими карбоновыми кислотами исследовано методом обращенно-фазной высокоэффективной жидкостной хроматографии. Подвижная фаза – ацетонитрил-вода, (80/20, по объему) с добавкой 0,1% муравьиной кислоты. Колонка – LiChrosorb RP 18, УФ детектор работал при длине волны λ = 254 нм и температуре 26°С. Определены основные хроматографические характеристики (время удерживания t_R и коэффициент емкости k') ароматических карбоновых кислот. Рассчитаны

значения липофильности log P кислот, а также значения констант связывания K_A (387-941 M^{-1}) и свободных энергий Гиббса ΔG (-14.74 – -16.94 kJ/mol) комплексов каликсарена с ароматическими карбоновыми кислотами. Молекулярное моделирование (Hyper Chem, версия 8.0) каликсареновых комплексов показало присутствие водородных связей между карбоксильными группами кислот и атомами азота имино-групп на верхнем ободе или атомами кислорода гидроксильных групп на нижнем ободе каликсаренового макроцикла. Оценено влияние липофильности log P кислот на значения K_A каликсареновых комплексов. Линейная зависимость констант связывания K_A от липофильности кислот свидетельствует о существенной роли сольвофобных взаимодействий в процессе комплексообразования. Установлена взаимосвязь между супрамолекулярными (K_A) и физико-химическими характеристиками (молекулярная масса, log P) кислот. Константы связывания K_A комплексов растут с увеличением молекулярной массы кислот и повышением значений их липофильности log P.

An important problem in chemistry and biology is molecular recognition, separation, membrane transport and analytical sensing of biorelevant molecules by artificial receptors [1-7]. Calixarenes – "macrocyclic vases", which are easily available through the cyclocondensation of *para*-substituted phenols with formaldehyde, – are widely used as molecular platforms for constructing specific receptors capable of highly selective recognition between fairly similar substrates [8-10]. Apparently, the outstanding receptor properties of functionalized calixarenes toward the biorelevant molecules make them highly promising materials for sensor technologies [11], as well as Host molecules for drug delivery systems in pharmaceutical science [5, 6, 8, 12-17].

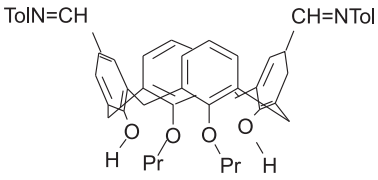
Aromatic carboxylic acids, such as benzoic, *p*-coumaric, cinnamic, gallic, diphenylacetic acid and their different derivatives are used in medical practice as antibacterial and antifungal agents for skin diseases and mycosis [18-23]. Many naturally occurring phenolic acids and analogues, namely caffeic and gallic acids, are known to exhibit a wide variety of biological functions, in addition to their primary antioxidant activity, which are mainly related to modulation of carcinogenesis [24].

The information on the supramolecular Host-Guest interaction of calix[4]arenes with the aromatic carboxylic acids will be useful in the design of artificial receptors for such biorelevant compounds.

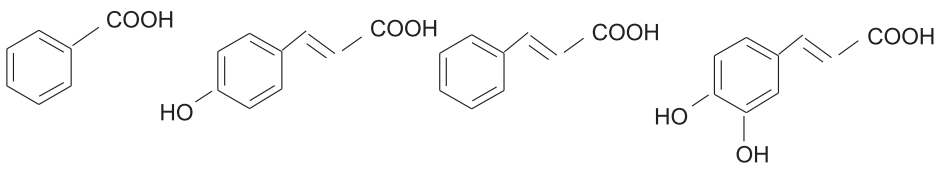
In this paper we reported the Host-Guests complexation study of 5,17-bis-(N-tolyliminomethyl)-25,27-dipropoxycalix[4]arene (**DPCA**) with benzoic **1**, *p*-coumaric **2**, cinnamic **3**, caffeic **4,** gallic **5**, diphenylacetic acid **6** (Scheme) by the reversed-phase high-performance liquid chromatography (RP HPLC) method in acetonitrile-water solution.

Experimental Part

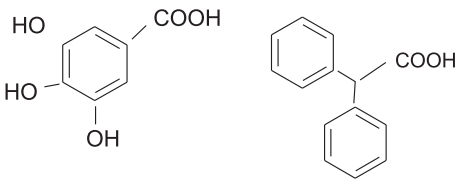
The RP HPLC study was performed on a Hitachi chromatograph (Hitachi, Ltd., Tokyo, Japan) consisting of a high-pressure pump connected to a Rheodyne sample 7120 injector with a 20 μL loop (Rheodyne,



DPCA



1 **2** **3** **4**



5 6

Scheme

4

Table

Retention times t_R, capacity factors *k'* of carboxylic acids **1-6**, K_A and ΔG values of their complexes with **DPCA**

Substrate	Retention time, t_R, min	Capacity factor, *k'*	K_A, M^{-1}	ΔG[a], kJ/mol
1[b]	4.50	0.50	650±72	-16.02
2	3.68	0.23	692±111	-16.18
3	3.80	0.27	941±175	-16.94
4	3.90	0.30	520±70	-15.47
5	4.0	0.33	625±88	-15.92
6	4.48	0.49	387±48	-14.74

[a] ΔG = -RT ln K_A
[b] K_A was determined in [28]

Berkeley, USA) and an ultraviolet-visible detector. The column (250×4.6 mm i.d.) was packed with Li-Chrosorb RP 18 (Merck, Darmstadt, Germany). Acetonitrile was bought from the Acros Organics. Carboxylic acids were purchased from Sigma-Aldrich (St. Louis, MO, USA). **DPCA** was synthesized by the method [25]. The acetonitrile-water (80/20, v/v) mixture was used as a blank mobile phase. The calixarene based mobile phases were prepared by dissolving **DPCA** in acetonitrile-water (80/20, v/v), 0.1% formic acid mixture to obtain the **DPCA** concentration of 0.05-0.6 mM. The analytes for injections were dissolved in the mixture of acetonitrile-water (80/20, v/v) (C = 0.01 mM). The amount of the sample injected was 20 μL. All chromatograms were recorded at 26°C. The UV detector operated at 254 nm. The dead time t_0 was measured with $NaNO_2$.

Determination of lipophilicity of log P of acids 1-6

Lipophilicity of *log P* of acids **1-6** (Table) was calculated by the HPLC method from equation *log P* = K x (log *k'*). The coefficient K being the relationship of *log P* value of benzoic acid **1** (1.87) [26] to its log *k'* was determined by RP HPLC in this work.

Molecular modelling

Molecular modelling of **DPCA** complexes with acids **1-6** were carried out using a Hyper Chem, version 8.0 programme [27]. The structures were optimized by the semi-empirical PM3 method.

Results and Discussion

Calixarene **DPCA** and carboxylic acids **1-6** in the given conditions of analysis were registered on the chromatograms as sharp peaks. The chromatographic characteristics of carboxylic acids **1-6** (retention time t_R, capacity factor *k')*, their binding constants K_A and free Gibbs energies ΔG of their complexes with **DPCA** are presented in Table.

Binding constants of the inclusion Host-Guest complexes of **DPCA** with aromatic carboxylic acids **1-6** were determined by the RP HPLC method described in [29] and based on determination of retention factor *k'* of the Guest – carboxylic acids prior to and after the Host addition to the mobile phase. The **DPCA** addition to the mobile phase decreases retention factor *k'* of carboxylic acids **1-6**. The linear character plots of 1/*k'vs* the **DPCA** concentration (Fig. 1) indicate formation of the Host-Guest supramolecular complexes with 1:1 stoichiometry.

In accordance with the data obtained (Table) the highest K_A was observed for cinnamic acid (941 M^{-1}), and the lowest K_A was observed for the most bulky diphenylacetic acid (387 M^{-1}). The binding constants K_A strongly depended on the molecular weight (Fig. 2) and lipophilicity of *log P* (Fig. 3) of the acids.

There is the linear dependence of the binding constants K_A on lipophilicity of *log P* of cinnamic, *p*-coumaric, gallic, caffeic, benzoic and diphenylacetic acid (Fig. 3).

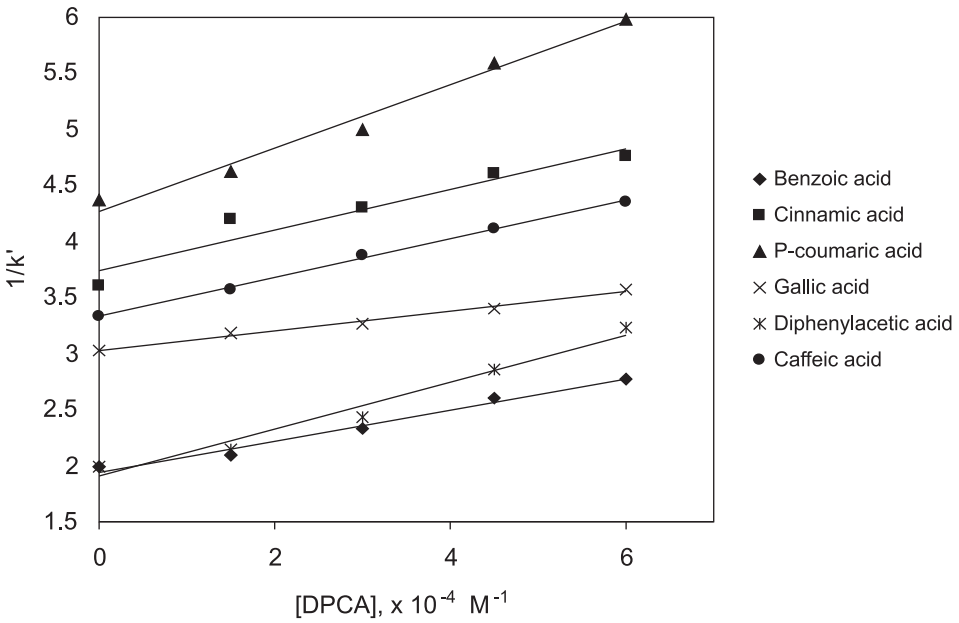


Fig. 1. Plots of 1/*k'vs* the **DPCA** concentration (r = 0.98-0.99).

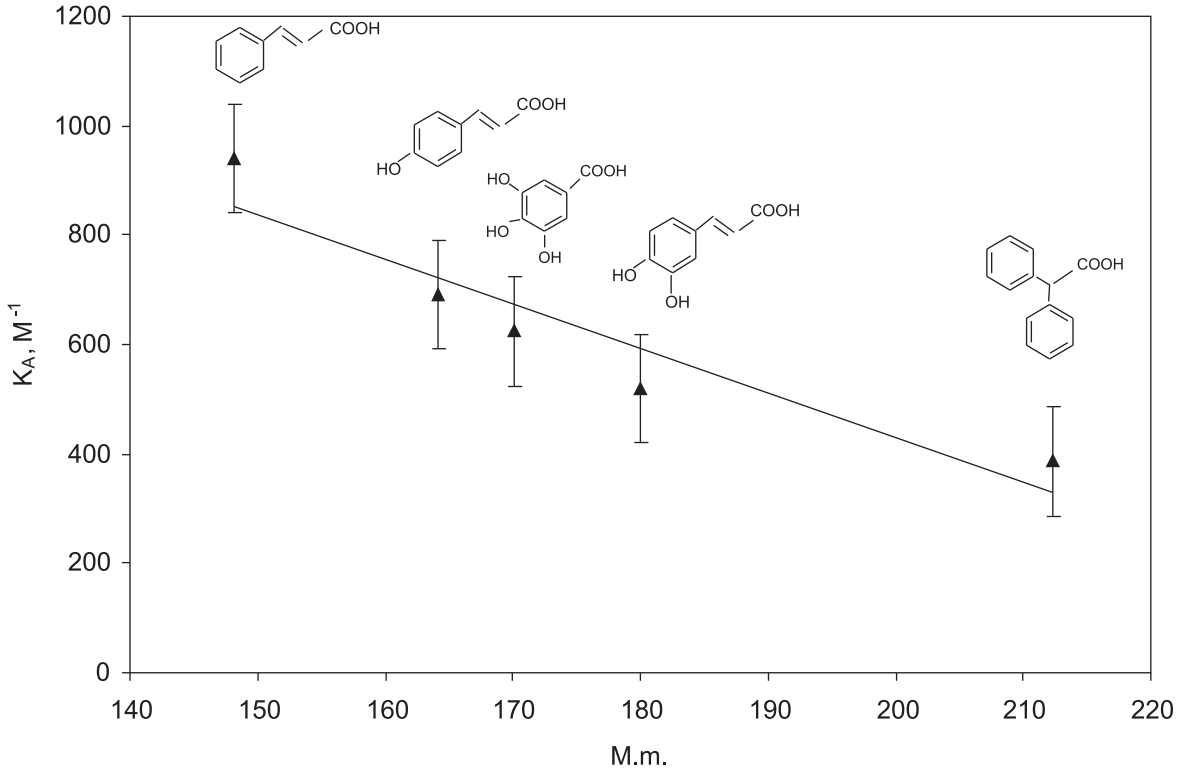


Fig. 2. The influence of the molecular weight of cinnamic, *p*-coumaric, gallic, caffeic and diphenylacetic acids on K_A of their complexes (r = 0.98).

The increase of *log P* values of the acids leads to increase of K_A values of their complexes with **DPCA**.

To clarify the nature of the supramolecular Host-Guest interactions the molecular modelling of **DPAA** complexes with cinnamic acid and diphenylacetic acid was carried out (Fig. 4).

Carboxylic acids deeply penetrate in the calixarene cavity (Fig. 4) with formation of the supramolecular Host-Guest complexes. The complexes are stabilized by the intermolecular hydrogen bonds C(O)O-H···OH formed by carboxylic groups of the Guest molecule with the oxygen atoms of the hydroxyl groups of the Host molecule.

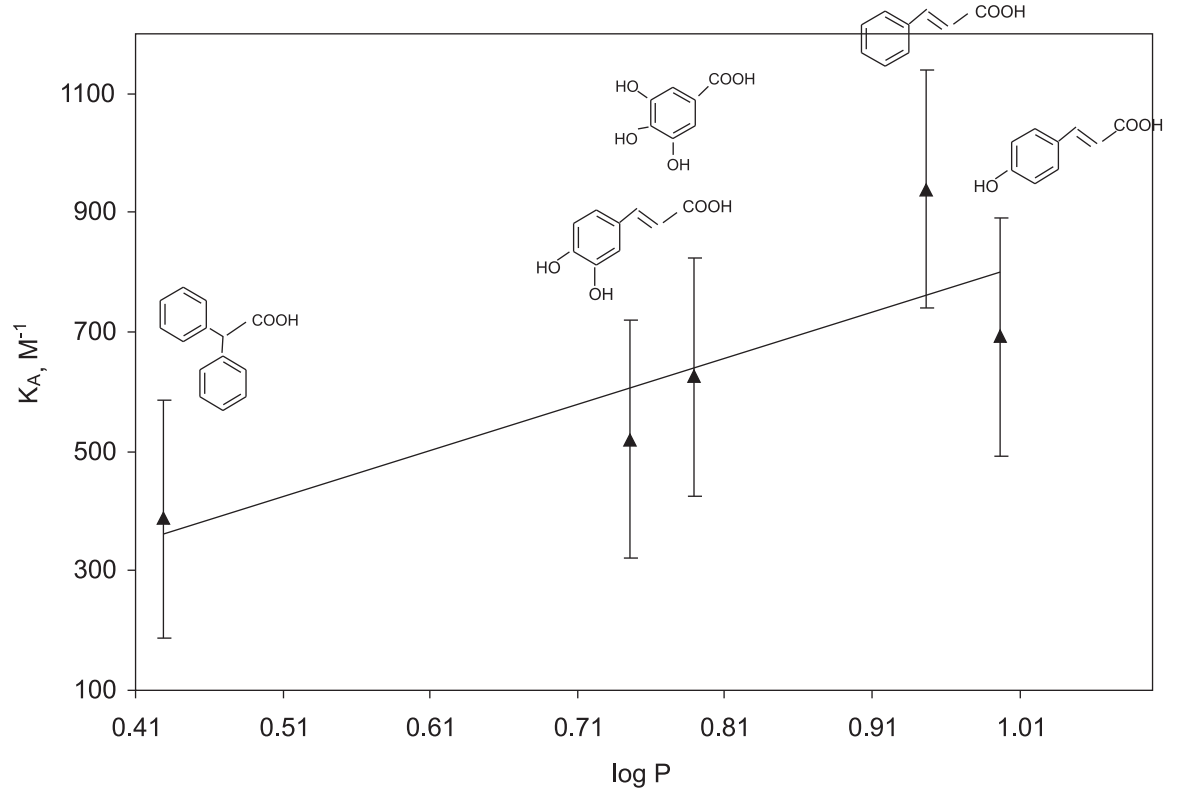


Fig. 3. Plots of K_A *vs log P* for diphenylacetic, caffeic, gallic, cinnamic and *p*-coumaric acids (r = 0.83).

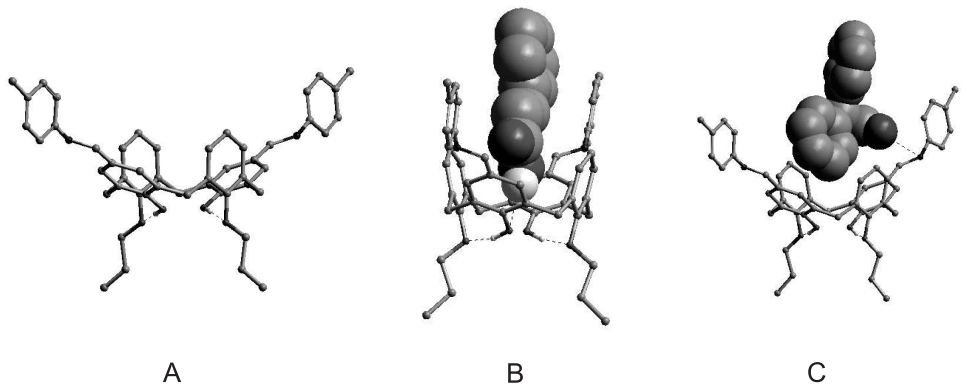


Fig. 4. The lowest energy structures of **DPCA (A)** and its complex with cinnamic (**B**) and diphenylacetic acid **(C)**. Intermolecular and intramolecular hydrogen bonds are presented by dotted lines.

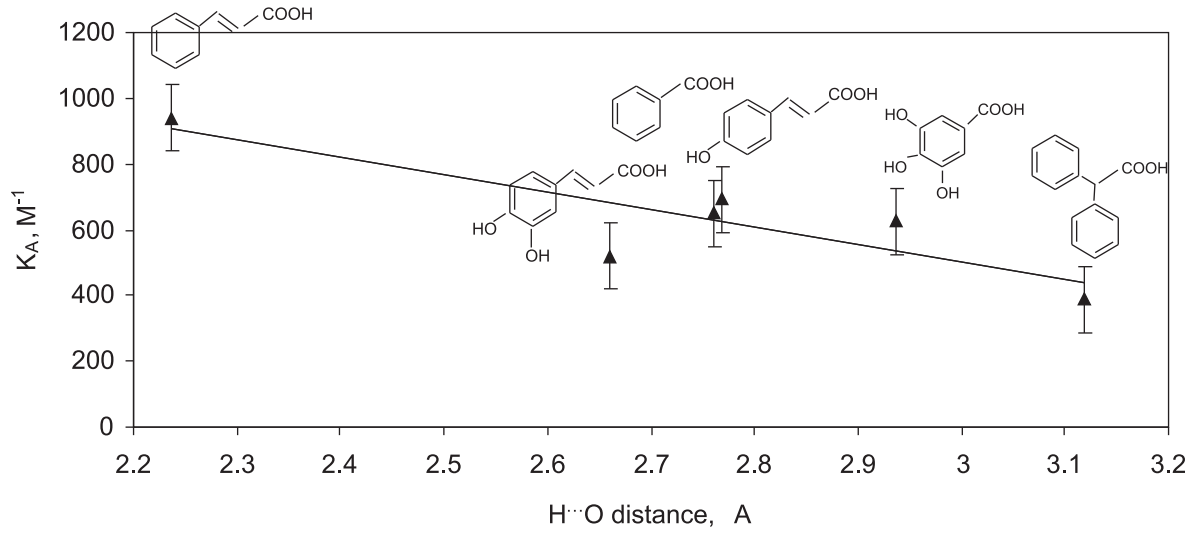


Fig. 5. Plots of K_A values *vs* H···O distances of the intermolecular hydrogen bonds between the Guest carboxylic groups and the Host hydroxyl groups at the lower rim of the macrocycle for cinnamic, caffeic, benzoic, *p*-coumaric, gallic and diphenylacetic acids (r = 0.86).

The proportional dependence of the binding constants of the complexes on the long hydrogen bonds H···O is observed (Fig. 5). Additionally, the complexes can be stabilized by the van der Waals stacking and interactions between the Host and Guest molecules (Fig. 4). In the case of diphenylacetic and benzoic acids the other hydrogen bonding is observed. Carboxylic groups form intermolecular bonds with the basic nitrogen atoms of imino groups (Fig. 4C). Phenyl groups of these acids are included into the molecular cavity as a result of π-π-stacking interactions. Plots of K_A values *vs* H···O distances of the intermolecular hydrogen bonds between the Guest carboxylic groups and the Host hydroxyl groups at the lower rim of the macrocycle is presented in Fig. 5.

Conclusions

DPCA containing two imino groups at the upper rim of the macrocycle forms the Host-Guest inclusion complexes with biorelevant aromatic carboxylic acids. Their binding constants (387-941 M^{-1}) in acetonitrile-water solution depend on the molecular weight and log P of the acids. The complexes are stabilized by intermolecular hydrogen bonds between the Guest carboxylic groups and the Host hydroxyl groups, van der Waals stacking and solvophobic interactions. Calixarene is a promising compound in the design of sensor devices or drug delivery systems for such biorelevant compounds.

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THE SYNTHESIS AND ANALGESIC PROPERTIES OF *N*-(BENZYL)-2-HYDROXY-9-METHYL-4-OXO-4*H*-PYRIDO[1,2-*a*]PYRIMIDINE-3-CARBOXAMIDES

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Key words: amidation; tricarbonylmethane heterocyclic derivatives; 2-hydroxy-4-oxo-4H-pyrido[1,2-a] pyrimidine-3-carboxamides; synthesis; bioisosteric replacements; analgesic activity

Continuing the search for new analgesics among derivatives of azahetarylcaboxylic acids by the reaction of ethyl 2-hydroxy-9-methyl-4-oxo-4H-pyrido[1,2-a]pyrimidine-3-carboxylate and benzylamines in boiling ethanol the corresponding group of N-(benzyl)-2-hydroxy-9-methyl-4-oxo-4H-pyrido[1,2-a]pyrimidine-3-carboxamides has been synthesized. The structure of the compounds obtained has been confirmed by the data of elemental analysis and NMR ^{1}H spectroscopy. It is noted that the signals of aromatic protons of pyrido-pyrimidine nuclei are shifted downfield and generally form a typical AMX spin system. At the same time, the signals of aromatic protons of benzilamide fragments on the contrary are shifted upfield in all cases and focused on very narrow segments of the spectra, thereby undergoing strong distortion. According to the results of the primary pharmacological screening it has been found that using the standard model of "acetic acid writhings" all N-(benzyl)-2-hydroxy-9-methyl-4-oxo-4H-pyrido[1,2-a]pyrimidine-3-carboxamides without exception have analgesic properties to a greater or lesser degree. Practically the same regularities of the benzylamide fragment structure –biological effect relationship as in the case of 4-hydroxyquinolin-2-ones analogues have been found. Based on it the conclusion about bioisosterism of 4-hydroxyquinolin-2-one and 2-hydroxy-9-methyl-4-oxo-4H-pyrido[1,2-a]pyrimidine nuclei has been made.

СИНТЕЗ ТА АНАЛГЕТИЧНІ ВЛАСТИВОСТІ N-(БЕНЗИЛ)-2-ГІДРОКСИ-9-МЕТИЛ-4-ОКСО-4Н-ПІРИДО[1,2-а] ПІРИМІДИН-3-КАРБОКСАМІДІВ

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Ключові слова: *амідування; гетероциклічні похідні трикарбонілметану; 2-гідрокси-4-оксо-4Н-піридо [1,2-а]піримідин-3-карбоксаміди; синтез; біоізостеричні переміщення; аналгетична активність*

Продовжуючи пошук нових аналгетиків серед похідних азагетарилкарбонових кислот, реакцією етил 2-гідрокси-9-метил-4-оксо-4Н-піридо[1,2-а]піримідин-3-карбоксилату з бензиламінами у киплячому етанолі ми здійснили синтез групи відповідних N-(бензил)-2-гідрокси-9-метил-4-оксо-4Н-піридо[1,2-а]піримідин-3-карбоксамідів. Для підтвердження будови одержаних речовин використані елементний аналіз та спектроскопія ^{1}Н ЯМР. Помічено, що сигнали ароматичних протонів піридо-піримідинового ядра зсунуті у слабке поле і в цілому утворюють типову AMX спінову систему. В той же час сигнали ароматичних протонів бензиламідних фрагментів навпаки в усіх випадках зміщені у відносно сильне поле та зосереджені на дуже вузьких відрізках спектрів, за рахунок чого піддаються досить сильному спотворенню. За результатами первинного фармакологічного скринінгу встановлено, що на стандартній моделі оцтовокислих «корчів» всі без виключення N-(бензил)-2-гідрокси-9-метил-4-оксо-4Н-піридо[1,2-а]піримідин-3-карбоксаміди в тій чи іншій мірі виявляють аналгетичні властивості. При цьому знайдені практично ті ж закономірності впливу будови бензиламідного фрагмента на біологічний ефект, що й у випадку 4-гідроксихінолін-2-онових аналогів. На підставі цього зроблено висновок щодо біоізостерності 4-гідроксихінолін-2-онового та 2-гідрокси-9-метил-4-оксо-4Н-піридо[1,2-а]піримідинового ядер.

СИНТЕЗ И АНАЛЬГЕТИЧЕСКИЕ СВОЙСТВА N-(БЕНЗИЛ)-2-ГИДРОКСИ-9-МЕТИЛ-4-ОКСО-4Н-ПИРИДО [1,2-а]ПИРИМИДИН-3-КАРБОКСАМИДОВ

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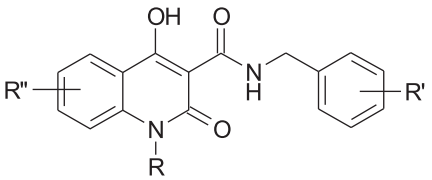
Ключевые слова: *амидирование; гетероциклические производные трикарбонилметана; 2-гидрокси-4-оксо-4Н-пиридо[1,2-а]пиримидин-3-карбоксамиды; синтез; биоизостерические перемещения; анальгетическая активность*

Продолжая поиск новых анальгетиков среди производных азагетарилкарбоновых кислот, реакцией этил 2-гидрокси-9-метил-4-оксо-4Н-пиридо[1,2-а]пиримидин-3-карбоксилата с бензиламинами в кипящем этаноле мы осуществили синтез группы соответствующих N-(бензил)-2-гидрокси-9-метил-4-оксо-4Н-пиридо[1,2-а] пиримидин-3-карбоксамидов. Для подтверждения строения полученных веществ использованы элементный анализ и спектроскопия ^{1}Н ЯМР. Отмечено, что сигналы ароматических протонов пиридо-пиримидинового ядра сдвинуты в слабое поле и в целом образуют типичную AMX спиновую систему. В то же время сигналы ароматических протонов бензиламидных фрагментов наоборот во всех случаях смещены в относительно сильное поле и сосредоточены на очень узких отрезках спектров, за счет чего претерпевают сильное искажение. По результатам первичного фармакологического скрининга установлено, что на стандартной модели уксуснокислых «корчей» все без исключения N-(бензил)-2-гидрокси-9-метил-4-оксо-4Н-пиридо[1,2-а]пиримидин-3-карбоксамиды в той или иной степени обладают анальгетическими свойствами. При этом обнаружены практически те же закономерности влияния строения бензиламидного фрагмента на биологический эффект, что и в случае 4-гидроксихинолин-2-оновых аналогов. На этом основании сделан вывод о биоизостерности 4-гидроксихинолин-2-онового и 2-гидрокси-9-метил-4-оксо-4Н-пиридо[1,2-а]пиримидинового ядер.

Among the world population it is almost impossible to find someone who would be unfamiliar with frightening and something paradoxical sensation of pain. On the one hand, the pain in its nature has a specific and necessary role for the survival – it instantly signals the occurrence of external or internal factors that can cause harm to the body. However, on the other hand, pain is able to exhaust the resources of the body very quickly and lead to serious disorders of its vital functions. This explains why the mankind is searching for means of dealing with pain throughout the history of its existence, and the desire to create an "ideal analgesic", which would meet all modern requirements for efficacy and safety, have not lost their relevance today [1-6].

The interesting objects of study in this respect are the numerous *N*-(benzyl)-4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxamides of the general formula (**1**) and their heterocyclic analogues studied earlier. Among them promising lead compounds far exceeding the analgesic effect of narcotic analgesics officially recognized and with a much lower toxicity have been identified [7-10].



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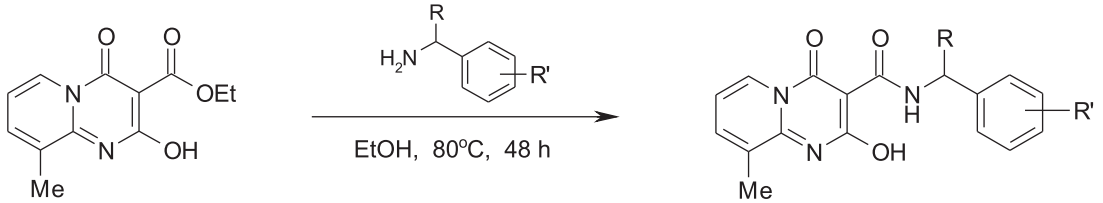
Continuing research in this area we considered replacement of the 4-hydroxyquinoline-2-ones base with 2-hydroxy-4-oxopyrido[1,2-*a*]pyrimidine nucleus that is similar by its structure as one of the possible ways to optimize analgesics of formula (**1**). The theoretical precondition for such modification was the methodology of bioisosteric replacements widely and effectively used by modern medical chemistry, involving the replacement of one group in the molecule close to it by the properties [11, 12]. It should be remembered that the bioisosteric groups are groups that are the same not only in size or volume, but have similar physical and chemical properties, and therefore, reveal a similar pharmacological effect [13-15]. In other words, the structural similarity of 4-hydroxyquinoline-2-ones and 2-hydroxy-4-oxopyrido[1,2-*a*] pyrimidine heterocycles itself does not guarantee their bioisosterism. Only the experimental study will show whether the compounds synthesized can exhibit the real analgesic effect.

The synthesis of the objects of research was carried out by the reaction of ethyl 2-hydroxy-9-methyl-4-oxo-4H-pyrido[1,2-*a*]pyrimidine-3-carboxylate (**2**) and benzylamines in boiling ethanol. As known, pyrido[1,2-*a*]pyrimidine esters of type (**2**) not only form rather stable salts with the primary amines, but also lose much in the reactivity [16]. Therefore, for successful amidation it is necessary to introduce at least a double excess of amine in the synthesis and significantly increase duration of the reaction.

It is interesting to note that in contrast to the initial ester (**2**) *N*-(benzyl)-4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxamides (**3a-n**) obtained do not form stable salts with the excess of the corresponding amine in the reaction mixture (apparently due to decrease of the 2-OH-group acidity [16]), and no additional procedures are required for their isolation.

All *N*-(benzyl)-4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxamides (**3a-n**) synthesized are colourless crystals with a sharp melting points (Table 1), moderately soluble in DMF and DMSO at room temperature, and practically insoluble in water. To confirm their structure the data of the elemental analysis and ^{1}H NMR spectroscopy were used (Table 2).

As a characteristic feature of the ^{1}H NMR spectra of N-(benzyl)-4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxamides (**3a-n**) it should be noted a great shift downfield the doublet signals of aromatic protons in position 6 of the pyrido-pyrimidine ring; it is caused by the proximity with a cyclic nitrogen atom. Their nearest neighbours – H-7 and H-8 – also resonate in the strongly (about 0.7 ppm) distinct areas, generally forming a typical AMX spin system (see Fig.). At the same time, the signals of aromatic protons of benzylamide fragments are shifted in a relatively strong field in all cases and focused on very narrow segments of the spectra, thereby undergoing strong distortion (see, for example, almost a singlet signal of the protons of H-5 'and H-6' piperonylamide **3k**).



2 **3a-n**

2: R = H – **a** R' = H; **b** R' = 4-F; **c** R' = 2-Cl; **d** R' = 4-Cl; **e** R' = 2-Me; **f** R' = 3-Me; **g** R' = 4-Me; **h** R' = 2-OMe; **i** R' = 4-OMe; **j** R' = 3,4-$(OMe)_2$; **k** R' = 3-O-CH_2-O-4. R = Me – **l** R' = H, (±); **m** R' = H, *S*(–); **n** R' = H, *R*(+)

Scheme

Table 1

Characteristics of benzylamides **3a-n**

Compound	Empirical formula	Found, % / Calculated, %			Mp, °C	Yield, %
		C	H	N		
3a	$C_{17}H_{15}N_3O_3$	65.94 / 66.01	4.77 / 4.89	13.62 / 13.58	161-163	89
3b	$C_{17}H_{14}FN_3O_3$	62.30 / 62.38	4.24 / 4.31	12.76 / 12.84	190-192	90
3c	$C_{17}H_{14}ClN_3O_3$	59.31 / 59.40	4.15 / 4.10	12.30 / 12.22	187-189	86
3d	$C_{17}H_{14}ClN_3O_3$	59.46 / 59.40	4.17 / 4.10	12.14 / 12.22	196-198	92
3e	$C_{18}H_{17}N_3O_3$	66.93 / 66.86	5.25 / 5.30	12.91 / 13.00	176-178	86
3f	$C_{18}H_{17}N_3O_3$	66.95 / 66.86	5.34 / 5.30	12.95 / 13.00	133-135	90
3g	$C_{18}H_{17}N_3O_3$	66.78 / 66.86	5.37 / 5.30	13.06 / 13.00	162-164	91
3h	$C_{18}H_{17}N_3O_4$	63.80 / 63.71	4.97 / 5.05	12.29 / 12.38	200-202	83
3i	$C_{18}H_{17}N_3O_4$	63.78 / 63.71	5.03 / 5.05	12.44 / 12.38	171-173	87
3j	$C_{19}H_{19}N_3O_5$	61.70 / 61.78	5.26 / 5.18	11.31 / 11.38	168-170	88
3k	$C_{18}H_{15}N_3O_5$	61.11 / 61.19	4.35 / 4.28	11.78 / 11.89	195-197	91
3l	$C_{18}H_{17}N_3O_3$	66.94 / 66.86	5.39 / 5.30	12.91 / 13.00	141-144	81
3m*	$C_{18}H_{17}N_3O_3$	66.95 / 66.86	5.57 / 5.30	12.94 / 13.00	144-146	84
3n**	$C_{18}H_{17}N_3O_3$	66.78 / 66.86	5.24 / 5.30	13.08 / 13.00	144-146	82

* $[\alpha]^{20}_D$ = + 23.2; *c* = 5; DMF.
** $[\alpha]^{20}_D$ = – 23.2; *c* = 5; DMF.

All biological experiments were carried out in full accordance with the European Convention on the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes and the Ukrainian Law No. 3447-IV "On protection of animals from severe treatment" (2006).

The analgesic activity of the compounds synthesized was studied on nonlinear white mice weighing 18-23 g (10 animals per each substance tested) using the standard model of "acetic acid writhings" [17]. The nociceptive effect was reproduced by intraperitoneal injection of 0.6% acetic acid solution in the amount of 0.1 ml per 10 g of the body weight 1 hour after administration of the test sample. The animals were observed for 20 min, counting the number of "writhings". The analgesic effect was assessed by the

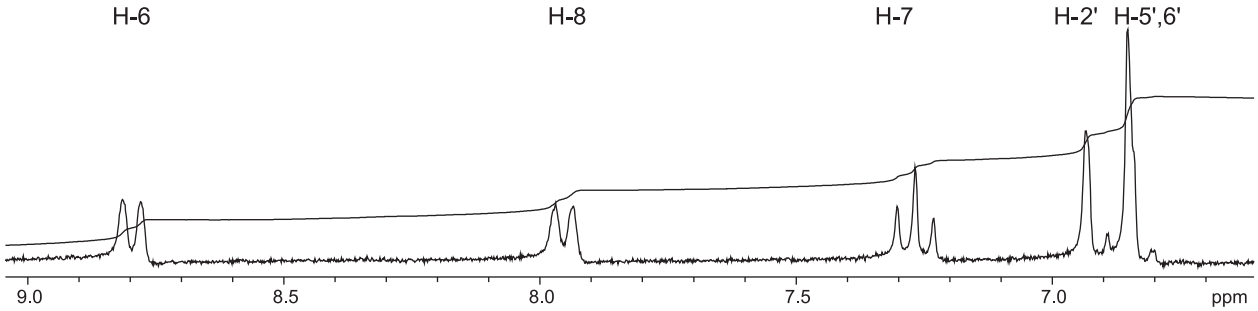


Fig. The fragment of ^{1}H NMR spectra (signals of aromatic protons) of amide **3k**.

Table 2

^{1}H NMR Spectra of benzylamides **3a-n**

Compound	Chemical shifts, δ, ppm (*J*, Hz)
3a	15.89 (1H, s, 2-OH); 9.97 (1H, t, *J* = 5.9, NH); 8.82 (1H, d, *J* = 7.0, H-6); 7.97 (1H, d, *J* = 7.0, H-8); 7.39-7.33 (5H, m, Ph); 7.28 (1H, t, *J* = 7.0, H-7); 4.60 (2H, d, *J* = 5.8, CONHC$\underline{H}_2$); 2.43 (3H, s, 9-Me)
3b	15.84 (1H, s, 2-OH); 9.96 (1H, t, *J* = 5.8, NH); 8.80 (1H, d, *J* = 7.0, H-6); 7.96 (1H, d, *J* = 7.0, H-8); 7.40 (2H, dd, *J* = 7.7 and 5.4, H-3′,5′); 7.27 (1H, t, *J* = 7.1, H-7); 7.16 (2H, t, *J* = 8.9, H-2′,6′); 4.57 (2H, d, *J* = 5.8, CONHC$\underline{H}_2$); 2.41 (3H, s, 9-Me)
3c	15.68 (1H, s, 2-OH); 10.03 (1H, t, *J* = 6.0, NH); 8.83 (1H, d, *J* = 7.1, H-6); 7.97 (1H, d, *J* = 6,9, H-8); 7.51-7.24 (5H, m, H-7 + H-3′,4′,5′,6′); 4.66 (2H, d, *J* = 6.2, CONHC$\underline{H}_2$); 2.42 (3H, s, 9-Me)
3d	15.82 (1H, s, 2-OH); 9.99 (1H, t, *J* = 6.0, NH); 8.81 (1H, d, *J* = 7.0, H-6); 7.97 (1H, d, *J* = 6.9, H-8); 7.41-7.37 (4H, m, H-2′,3′,5′,6′); 7.28 (1H, t, *J* = 7.0, H-7); 4.58 (2H, d, *J* = 6.0, CONHC$\underline{H}_2$); 2.42 (3H, s, 9-Me)
3e	15.82 (1H, s, 2-OH); 9.85 (1H, t, *J* = 5.8, NH); 8.77 (1H, d, *J* = 6,9, H-6); 7.93 (1H, d, *J* = 6.9, H-8); 7.29-7.10 (5H, m, H-7 + H-3′,4′,5′,6′); 4.57 (2H, d, *J* = 5.8, CONHC$\underline{H}_2$); 2.40 (3H, s, 9-Me); 2.31 (3H, s, 2′-Me)
3f	15.91 (1H, s, 2-OH); 9.94 (1H, t, *J* = 6.1, NH); 8.80 (1H, d, *J* = 7.1, H-6); 7.95 (1H, d, *J* = 7.1, H-8); 7.32-7.04 (5H, m, H-7 + H-2′,4′,5′,6′); 4.55 (2H, d, *J* = 6.1, CONHC$\underline{H}_2$); 2.41 (3H, s, 9-Me); 2.28 (3H, s, 3′-Me)
3g	15,88 (1H, s, 2-OH); 9,87 (1H, t, *J* = 5,8, NH); 8,76 (1H, d, *J* = 7,1, H-6); 7,93 (1H, d, *J* = 7,1, H-8); 7,25 (1H, t, *J* = 6,9, H-7); 7,20 (2H, d, *J* = 8,0, H-2′,6′); 7,13 (2H, d, *J* = 8,0, H-7 + H-3′,5′); 4,52 (2H, d, *J* = 5,9, CONHC$\underline{H}_2$); 2,39 (3H, s, 9-Me); 2,25 (3H, s, 4′-Me)
3h	15.93 (1H, s, 2-OH); 9.95 (1H, t, *J* = 6.0, NH); 8.81 (1H, d, *J* = 7.1, H-6); 7.95 (1H, d, *J* = 7.1, H-8); 7.33-7.21 (3H, m, H-7 + H-3′,5′); 7.03 (1H, d, *J* = 8.0, H-6′); 6.91 (1H, t, *J* = 7.6, H-4′); 4.54 (2H, d, *J* = 6.0, CONHC$\underline{H}_2$); 3.83 (3H, s, OMe); 2.41 (3H, s, 9-Me)
3i	15,94 (1H, s, 2-OH); 9,87 (1H, t, *J* = 5,8, NH); 8,78 (1H, d, *J* = 7,1, H-6); 7,95 (1H, d, *J* = 7,0, H-8); 7,32-7,20 (3H, m, H-7 + H-3′,5′); 6,89 (2H, d, *J* = 8,7, H-2′,6′); 4,50 (2H, d, *J* = 5,8, CONHC$\underline{H}_2$); 3,71 (3H, s, OMe); 2,41 (3H, s, 9-Me)
3j	15.96 (1H, s, 2-OH); 9.89 (1H, t, *J* = 6.0, NH); 8.80 (1H, d, *J* = 7,0, H-6); 7.95 (1H, d, *J* = 7.0, H-8); 7.27 (1H, t, *J* = 6.9, H-7); 6.99 (1H, s, H-2′); 6.92-6.87 (2H, m, H-5′,6′); 4.50 (2H, d, *J* = 6.0, CONHC$\underline{H}_2$); 3.73 (3H, s, OMe); 3.71 (3H, s, OMe); 2.42 (3H, s, 9-Me)
3k	15.90 (1H, s, 2-OH); 9.89 (1H, t, *J* = 6.0, NH); 8.80 (1H, d, *J* = 7.1, H-6); 7.95 (1H, d, *J* = 7.0, H-8); 7.27 (1H, t, *J* = 7.0, H-7); 6.93 (1H, s, H-2′); 6.88-6.82 (2H, m, H-5′,6′); 5.98 (2H, s, O-CH_2-O); 4.48 (2H, d, *J* = 6.0, CONHC$\underline{H}_2$); 2.40 (3H, s, 9-Me)
3l	15.73 (1H, s, 2-OH); 9.98 (1H, d, *J* = 7.3, NH); 8.82 (1H, d, *J* = 7.1, H-6); 7.96 (1H, d, *J* = 7.0, H-8); 7.44-7.28 (6H, m, H-7 + Ph); 5.17 (1H, q, *J* = 7.3, CONHC$\underline{H}$); 2.41 (3H, s, 9-Me); 1.53 (3H, d, *J* = 7.3, CH-$\underline{Me}$)
3m	15.73 (1H, s, 2-OH); 9.98 (1H, d, *J* = 7.3, NH); 8.82 (1H, d, *J* = 7.1, H-6); 7.96 (1H, d, *J* = 7.0, H-8); 7.44-7.28 (6H, m, H-7 + Ph); 5.17 (1H, q, *J* = 7.3, CONHC$\underline{H}$); 2.41 (3H, s, 9-Me); 1.53 (3H, d, *J* = 7.3, CH-$\underline{Me}$)
3n	15.73 (1H, s, 2-OH); 9.98 (1H, d, *J* = 7.3, NH); 8.82 (1H, d, *J* = 7.1, H-6); 7.96 (1H, d, *J* = 7.0, H-8); 7.44-7.28 (6H, m, H-7 + Ph); 5,17 (1H, q, *J* = 7.3, CONHC$\underline{H}$); 2.41 (3H, s, 9-Me); 1.53 (3H, d, *J* = 7.3, CH-$\underline{Me}$)

ability of compounds to reduce the number of "writhings" in the groups under study compared to the control and expressed in percentage (Table 3). Testing was carried out in comparison with such known non-narcotic analgesics as Piroxicam (Jenapharm, Germany), Diclofenac (KRK, Slovenia) and Nabumetone (Smith-Kline Beecham, Germany). All substances under study were administered orally in the screening dose of 20 mg/kg as a thin aqueous suspension stabilized with Tween-80. Medicines were used similarly or as aqueous solutions in the doses corresponding to their ED_{50} for this experimental model [18]. The animals of the control group received an equivalent amount of water and Tween-80. The results of all biological tests were statistically processed using the Student's *t*-test [19].

The analysis of the experimental data presented in Table 3 shows that the replacement of 4-hydroxyquinoline-2-one nucleus on the 2-hydroxy-4-oxopyrido[1,2-*a*]pyrimidine one was really bioisosteric since all compounds, without exception, revealed a more or less pronounced analgesic effect.

A comparative analysis with the parameters of the corresponding benzylamides of 1*H*- and 1-allylsubstituted 4-hydroxy-6,7-dimethoxy-2-oxo-1,2-dihydroquinoline-3-carboxylic acids studied under similar conditions shows approximately the same structural and biological regularities – the similar effect of the nature and location of substituents in the aromatic ring of the benzylamide moiety of the molecule can be traced in most of the examples (Table 3). Methylation of the methylene bridge separating phenyl and amide nitrogen – amides **3l-n** – regardless of the peculiarities of the spatial structure of the chiral fragment formed leads to the almost complete loss of analgesic properties, and therefore, it is undesirable. Of all groups of the compounds tested *N*-(benzyl)-

Table 3

The analgesic properties of benzylamides **3a-n**

Compound	Analgesic activity	
	Average number of "acetic acid writhing"	%*
3a	39.7±1.3	52.3 (66.6 & 30.9)
3b	56.9±2.2	31.5 (44.5 & 15.6)
3c	55.9±1.1	32.7 (45.5 & 14.4)
3d	40.6±1.4	51.2 (54.0 & 56.9)
3e	73.3±1.8	11.8
3f	68.8±1.6	17.1
3g	69.5±2.5	16.4 (12.4 & 24.3)
3h	60.9±1.2	26.7 (37.7 & 10.8)
3i	61.9±1.4	25.6 (36.5 & 26.3)
3j	47.8±1.0	42.4 (53.6 & 39.4)
3k	59.0±1.7	29.1 (53.4 & 15.4)
3l	68.1±1.3	18.2 (40.7 & 9.50)
3m	69.1±1.5	16.9
3n	68.6±1.1	17.5
Piroxicam (92 mg/kg)	41.6±1.8	50.0
Diclofenac (5 mg/kg)	40.1±2.3	51.6
Nabumetone (50 mg/kg)	41.0±3.3	50.6
Control	83.2±1.3	–

* In parentheses there are the indices of the analgesic activity of the corresponding benzylamides of 1*H*- and 1-allylsubstituted 4-hydroxy-6,7-dimethoxy-2-oxo-1,2-dihydroquinoline-3-carboxylic acids studied under similar conditions [20 & 9].

4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxamide (**3a**) and its 4-chloro-substituted analogue (**3d**) exhibiting the analgesic activity at the level of Piroxicam and Nabumetone but in much lower dose are of the most interest.

Experimental Part

The ^{1}H NMR spectra were recorded on a Varian Mercury-400 spectrometer (400 MHz) in DMSO-d_6 solution, the internal standard was TMS. Elemental analysis was carried out on a EuroVector EA-3000 microanalyzer. Melting points were determined in capillaries on a SMP10 Stuart digital melting point analyzer. The values of specific rotation of the optically active amides **3m,n** were determined on a Polamat A polarimeter. These syntheses use commercially *S(–)*- and *R(+)*-1-phenyl- and 1-(4-methoxyphenyl)ethylamines from Fluka with the optical purity of at least 99.5 and 99.0%, respectively. The starting ethyl 2-hydroxy-9-methyl-4-oxo-4*H*-pyrido[1,2-*a*]pyrimidine-3-carboxylate (**2**) was synthesized according to the literature procedure [21].

***N*-(Benzyl)-2-hydroxy-9-methyl-4-oxo-4H-pyrido[1,2-a]pyrimidine-3-carboxamides (3a-n). The general procedure.** Add the corresponding benzylamine (0.02 Mol) to 2.48 g (0.01 Mol) of the solution of ethyl ester (**2**) in 10 mL of ethanol and heat at reflux for 30 h. Then cool the reaction mixture and allow to stand at a temperature of about 0°C for 10-12 h. Filter the precipitate of benzylamide (3a-n) obtained, wash thoroughly with hexane, then with cold water and dry in the air. Crystallize from the DMF – ethanol mixture (1:1).

Conclusions

1. A new series of *N*-(benzyl)-2-hydroxy-9-methyl-4-oxo-4H-pyrido[1,2-*a*]pyrimidine-3-carboxamides has been synthesized as potential analgesics, their structure has been confirmed by elemental analysis and ^{1}H NMR spectra.

2. According to pharmacological studies it has been found that 2-hydroxy-9-methyl-4-oxopyrido[1,2-*a*]pyrimidine and 4-hydroxyquinoline-2-one are bioisosteric heterocycles and are of undoubted interest as the basis for obtaining highly effective analgesics.

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THE SYNTHESIS OF 4-THIAZOLIDINONE DERIVATIVES USING 2-(4-R-2-FORMYLPHENOXY)-N-(R'-PHENYL)ACETAMIDES AND THEIR ANTI-INFLAMMATORY ACTIVITY

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Key words: 4-thiazolidinones; NSAIDs; 2-(4-R-2-formylphenoxy)-N-(R'-phenyl)acetamides; anti-exudative activity

The research is devoted to the rational design of new non-steroidal anti-inflammatory drugs (NSAIDs) using the 4-thiazolidinone "core". A series of 2-(4-R-2-formylphenoxy)-N-(R'-phenyl)acetamides has been synthesized from salicylic aldehydes for structural modifications of basic heterocycles. The aldehydes obtained are active carbonyl agents and suitable "building blocks" for the focused synthesis of biologically active compounds. Ylidene derivatives of 2-thioxo-4-thiazolidinone and 2-(4-hydroxyphenyl)imino-4-thiazolidone have been synthesized in the Knoevenagel reaction conditions. The one-pot reaction between 3(5)-merkapto-1,2,4-triazoles, chloroacetic acid and the salicylic aldehyde derivatives synthesized have been used for the synthesis of 5-ylidene-thiazolo[3,2-b][1,2,4]triazol-6-one. Parameters of acute toxicity and the anti-exudative activity (carrageenin paw edema test) have been studied for the ylidene derivatives synthesized. It has been found that all compounds synthesized demonstrate the anti-exudative activity, and some "structure – acute toxicity – anti-exudative activity" relationships have been analyzed. Based on the results of in vivo studies the lead compound – 4-{2-[4-chloro-2-(6-oxothiazolo[3,2-b][1,2,4]triazole-5-ylidenemethyl)-phenoxy]-acetylamino}-benzoic acid ethyl ester that demonstrates the anti-exudative activity equivalent to the classic NSAID Diclofenac has been identified, it has a low level of toxicity and can be recommended for the profound study.

СИНТЕЗ ПОХІДНИХ 4-ТІАЗОЛІДИНОНУ З ВИКОРИСТАННЯМ 2-(4-R-2-ФОРМІЛФЕНОКСИ)-N-(R'-ФЕНІЛ) АЦЕТАМІДІВ ТА ЇХ ПРОТИЗАПАЛЬНА АКТИВНІСТЬ

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Ключові слова: *4-тіазолідинони; НПЗЗ; 2-(4-R-2-формілфенокси)-N-(R'-феніл)ацетаміди; антиексудативна активність*

Дослідження присвячено раціональному дизайну нових нестероїдних протизапальних лікарських засобів (НПЗЗ) з використанням 4-тіазолідинонового «каркасу». Для структурної модифікації цільового гетероциклу синтезовано ряд 2-(4-R-2-формілфенокси)-N-(R'-феніл)ацетамідів, які є активними карбонільними сполуками, зручними «building blocks» для спрямованого синтезу біологічно активних сполук. В умовах реакції Кньовенагеля з 2-тіоксо-4-тіазолідиноном, 2-(4-гідроксифеніл)іміно-4-тіазолідиноном та при однореакторній взаємодії з 3(5)-меркапто-1,2,4-триазолом і монохлороцтовою кислотою та синтезованими похідними саліцилових альдегідів отримано групу відповідних іліденпохідних. Для синтезованих іліденових похідних проведені дослідження параметрів гострої токсичності та антиексудативної активності з використанням карагенінової моделі запального процесу. Встановлено, що всі синтезовані сполуки демонструють антиексудативну активність та проаналізовані деякі закономірності «структура – гостра токсичність – антиексудативна активність». За результатами in vivo досліджень ідентифіковано сполуку – лідер – етиловий естер 4-{2-[4-хлоро-2-(6-оксотіазоло[3,2-b][1,2,4]триазол-5-ілідинметил)-феноксі]-ацетиламіно}-бензоатної кислоти, яка демонструє антиексудативну активність, еквівалентну лікарському засобу «Диклофенак» на фоні низької токсичності та може бути рекомендований для поглиблених досліджень.

СИНТЕЗ ПРОИЗВОДНЫХ 4-ТИАЗОЛИДИНОНА С ИСПОЛЬЗОВАНИЕМ 2-(4-R-2-ФОРМИЛФЕНОКСИ)-N-(R'-ФЕНИЛ)АЦЕТАМИДОВ И ИХ ПРОТИВОВОСПАЛИТЕЛЬНАЯ АКТИВНОСТЬ

С.М.Голота, К.Е.Данилюк, Т.И.Ющенко, Н.И.Волощук, О.В.Билык, Р.Б.Лесик

Ключевые слова: *4-тиазолидиноны; НПВС; 2-(4-R-2-формилфенокси)-N-(R'-фенил)ацетамиды; антиэкссудативная активность*

Исследование посвящено рациональному дизайну новых нестероидных противовоспалительных средств (НПВС) с использованием 4-тиазолидинонового «каркаса». Для структурной модификации целевого гетероцикла исходя из салициловых альдегидов синтезирован ряд 2-(4-R-2-формилфенокси)-N-(R'-фенил)ацетамидов, которые являются активными карбонильными соединениями и удобными «building blocks» для направленного синтеза биологически активных соединений. В условиях реакции Кнёвенагеля с 2-тиоксо-4-тиазолидиноном, 2-(4-гидроксифенил)имино-4-тиазолидиноном, а также при однореакторном взаимодействии 3(5)-меркапто-1,2,4-триазола, монохлоруксусной кислоты и синтезированных производных салициловых альдегидов получено группу соответствующих илиденпроизводных. Для синтезированных соединений проведены исследования параметров острой токсичности и антиэкссудативной активности с использованием каррагениновой модели воспалительного процесса. Установлено, что все синтезированные соединения демонстрируют противовоспалительное действие и проанализированы некоторые закономерности «структура – острая токсичность – антиэкссудативная активность». По результатам in vivo исследований идентифицировано соединение – лидер: этиловый эфир 4-{2-[4-хлор-2-(6-оксотиазоло[3,2-b][1,2,4]триазол-5-илиденметил)-феноксі]-ацетиламино}-бензойной кислоты, которое демонстрирует антиэкссудативную активность, эквивалентную лекарственному средству «Диклофенак» на фоне низкой токсичности и может быть рекомендовано для углубленных исследований.

Non-steroidal anti-inflammatory drugs (NSAIDs) are one of the oldest and most widely used groups of drugs today [1]. There are more than 20 original monocomponent NSAIDs, about 200 generics and a significant number of combined drugs and their generic versions at the Ukrainian pharmaceutical market [2]. However, the problem of searching and creating new NSAIDs remains open, primarily to eliminate gastrointestinal, cardiovascular and renovascular risks in their long-term use [3-5]. Drugs affect the cellular metabolism of arachidonic acid, which is a substrate for the synthesis of active intracellular intermediates, eicosanoids, leukotrienes and others, and is the key mechanism for therapeutic and adverse effects of different classes of NSAIDs [6]. Organic compounds from different classes [7-9], including 4-thiazolidinone derivatives [10-13], can change the phospholipase A2 activity blocking the process of arachidonic acid releasing from cell membranes phospholipids; selective and non-selective inhibit cyclooxygenases types 1 and 2 (COX-1 and COX-2) and thus prevent the arachidonic acid transformation to eicosanoids; inhibit 5-lipoxygenase (5-LOX) preventing the arachidonic acid conversion to leukotrienes; demonstrate multiactivity against the enzyme systems (COX-2/5-LOX dual inhibitors), etc. According to the modern concepts PPAR-receptors play an important role in the cellular mechanisms of inflammatory processes [14]. It is known that 4-thiazolidinones are "classic" high affinity ligands for PPARγ-receptors [15].

The systematic research in the field of potential NSAIDs synthesis and screening among 4-thiazolidinone derivatives is the priority direction for the research group of the Department of Pharmaceutical, Organic and Bioorganic Chemistry at Danylo Halytsky Lviv National Medical University [16-21]. The qualitative and quantitative "structure – antiinflammatory activity" databases obtained for 4-thiazolidinones allow to carry out the rational structural design of the 4-thiazolidinone "core" for searching new potential NSAIDs. The aim of this research was the synthesis of 5-ylidene-4-thiazolidinones from 2-(4-R-2-formylphenoxy)-N-(R'-phenyl)acetamides, as well as the study of their anti-exudative activity and acute toxicity.

Target 2-(4-R-2-formylphenoxy)-N-(R'-phenyl)acetamides **1-6** were obtained in the alkylation reaction of 2-hydroxy- and 5-chloro-2-hydroxybenzaldehydes (salicylic aldehydes) with chloroacetamide, N-(R'-phenyl)chloroacetamides and 2-chloro-1-[5-(4-methoxyphenyl)-3-phenyl-4,5-dihydropyrazol-1-yl]-ethanone in the ethanol medium in the presence of potassium hydroxide (Fig. 1).

Compounds **1-6** are useful "building blocks" for structural modification of 4-thiazolidinone scaffold in position 5, and they give the corresponding ylidene-derivatives with high yields in the Knoevenagel reaction with 2-thioxo-4-thiazolidinone (**7-9**, **11**, **13**), 2-(4-hydroxyphenyl)imino-4-thiazolidinone (**12**) and the one-pot reaction with 3(5)-mercapto-1,2,4-triazole and chloroacetic acid (**10**) (Fig. 2).

The structure of the compounds synthesized was confirmed by ^{1}H-NMR spectra. Protons of CH_2-CH fragments in the pyrazoline ring of compounds **6** and **13** form a characteristic AMX system due to their diastereoisomerism. This system appears in ^{1}H-NMR spectra as three duplicate doublets at 3.30-3.40, 4.00-4.15 and 5.70-5.90 ppm with constant J_{AM} = 17.8-18.0, J_{Ax} = 10.7-10.9 and J_{Mx} = 3.0-3.8 Hz.

The acute toxicity was studied in order to assess the prospects of the compounds synthesized as biologically active substances. Pastushenko's express – method was used for determination of acute toxicity parameters [22]. White mice of both sexes weighing 20-27 g were used for the experiment. The animals were kept on a standard diet with a free access to food and water during the experiment. The test compounds were dissolved in Tween-80 and purified water and introduced intraperitoneally. The observation of the animals was performed for 14 days. The LD_{50} values determined for the test substances (Fig. 3) were higher or equivalent to the LD_{50} of the reference drug Diclofenac and allowed to refer them to moderately

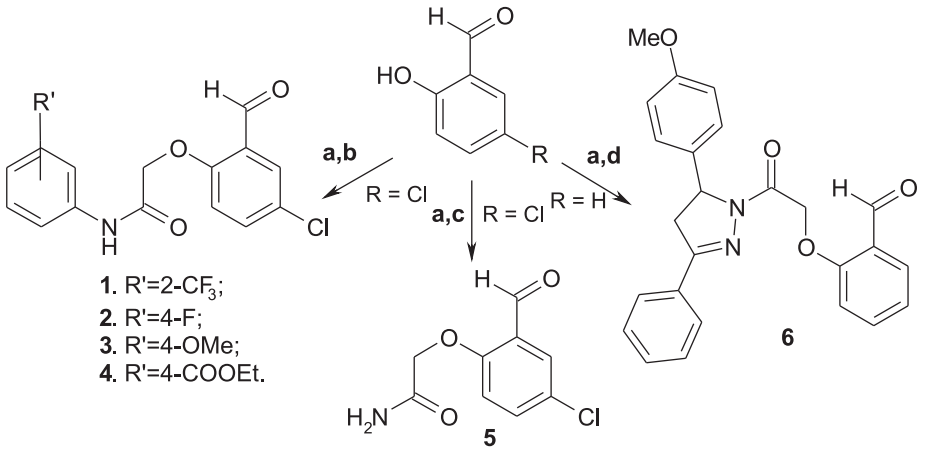

Fig. 1: **a** – KOH; **b** – N-(R'-phenyl)chloroacetamides; **c** – chloroacetamide; **d** – 2-chloro-1-[5-(4-methoxyphenyl)-3-phenyl-4,5-dihydropyrazol-1-yl]-ethanone.

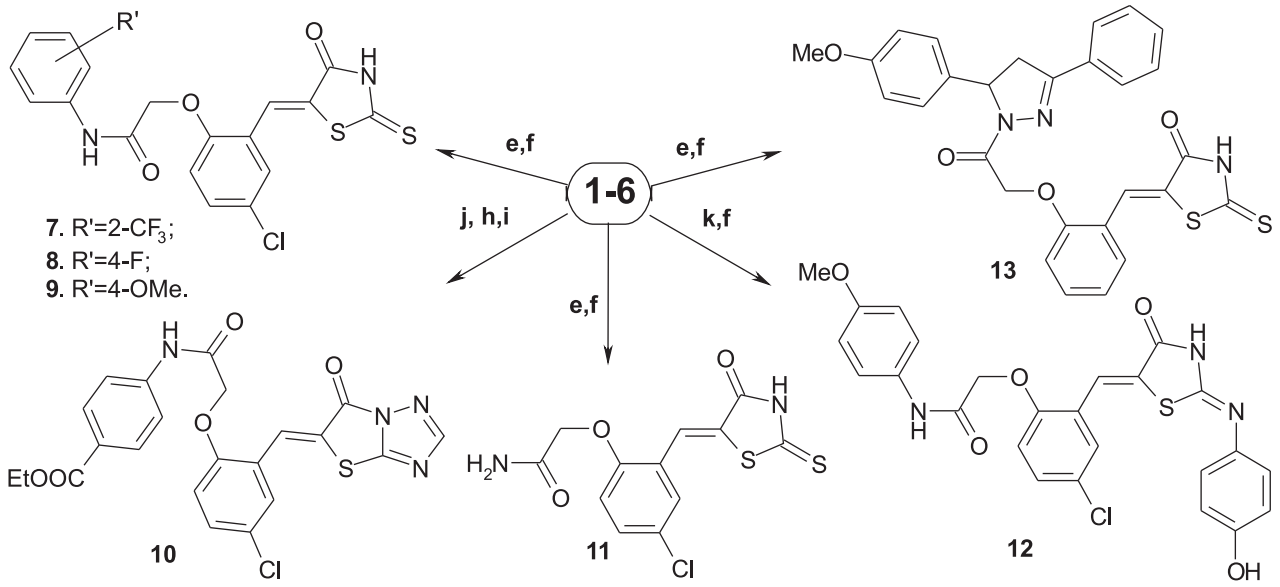

Fig. 2: **e** – 2-Thioxo-4-thiazolidinone; **f** – AcOH, AcONa; **j** – 3(5)-mercapto-1,2,4-triazole; **h** – $ClCH_2COOH$; **i** – Ac_2O, AcOH, AcONa; **k** – 2-(4-hydroxyphenyl)imino-4-thiazolidinone.

toxic (**7**) and low-toxic (**8-13**) compounds according to K. K. Sidorov classification (the III and IV class of toxicity) [23]. In the analysis of the "structure – acute toxicity" relationship it has been found that higher levels of LD_{50} have the following compounds: the annelated derivative of 4-thiazolidinone (**10**) containing the unsubstituted amide function (**11**) or the diphenylpyrazoline moiety (**13**) in the ylidene fragment, while derivatives **7-9**, **12** with N-(R'-phenyl)chloroacetamides substituents in the molecule are more toxic.

The carrageenin paw edema test was used for the anti-exudative activity screening of the compounds synthesized [24]. The inflammatory edema of the paw was generated by injection of 0.05 ml 1% carrageenin solution (Sigma) into the right hind limb of the mice [25]. The compounds and reference drug were administered intraperitoneally one hour before the carrageenin injection in the doses of 0.05 LD_{50}. The control group of animals was administered an equivalent amount of the solvent. Animals were taken out from the experiment by the cervical vertebrae dislocation at 3 hr after injecting carrageenan (at the peak of action) and limbs masses were measured and compared after disarticulation at the hip joints. The anti-exudative activity (AEA) of compounds was calculated using the equation:

$$\text{AEA} = 100\% - \frac{Me.e. - Mh.e.}{Me.c. - Mh.c.} \cdot 100\%,$$

where: *Me.e.(c) and Mh.e.(c)* are edema and healthy limbs weight for experimental and control animals, respectively.

The results of AEA screening (Fig. 3) demonstrate that all compounds synthesized are active. The range

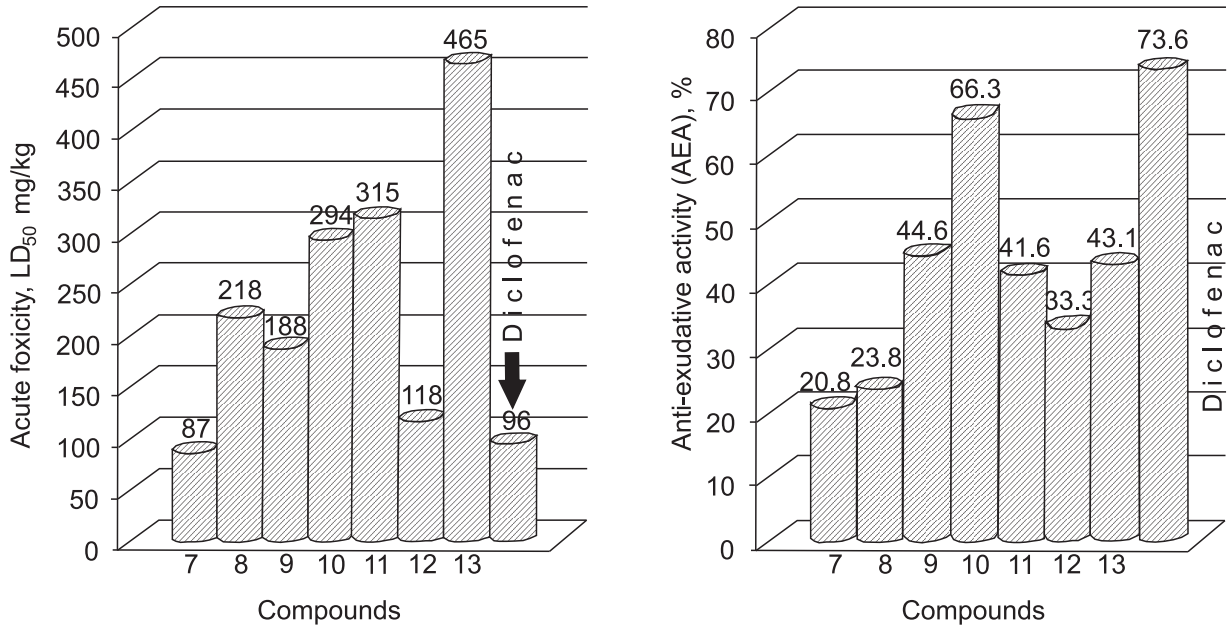

Fig. 3. Acute toxicity and the anti-exudative activity of compounds **7-13**.

of AEA values of the compounds synthesized in relation to the reference drug are from 27.2% (**7**) to 88.3% (**10**). Derivatives with a fluorine atom in the phenylacetamide fragment (**7**, AEA = 20.8%; **8**, AEA = 23.8%) are characterized by the lowest activity level. The change of fluorine to the methoxy group in the *p*-position of compound **8** causes a significant increase of the anti-inflammatory effect – **9**, AEA = 44.6%. The equivalent effect was obtained by simplifying the O-alkylmoiety in the ylidene fragment to the unsubstituted amide group (**11**, AEA = 41.6%), as well as by structural complication of the diphenylpyrazoline substituent (**13**, AEA = 43.1%). The presence of the *p*-hydroxyphenylimine fragment in position 2 of the 4-thiazolidinone core (**12**) causes decrease of AEA in relation to thioxoanalogue **9**, and is 33.3%. The anelated 4-thiazolidinone derivative **10** shows the highest AEA = 66.3% among the compounds synthesized and equivalent to the reference drug.

Experimental Part

Melting points were measured in open capillary tubes on a BŰCHI B-545 melting point apparatus and were uncorrected. The elemental analysis (C, H, N) was performed using a Perkin-Elmer 2400 CHN analyzer and was within ± 0.4% of the theoretical values. The ^{1}H-NMR spectra were recorded on a Varian Gemini spectrometer at 400 MHz using the mixture of DMSO-d_6+CCl_4 as a solvent and TMS as an internal standard. Chemical shift values are reported in ppm units with the use of δ scale.

***The general procedure for the synthesis of 2-(4-R-2-formylphenoxy)-N-(R'-phenyl)acetamides* 1-6**. Reflux the mixture of 5-chloro-2-hydroxy- (**1-5**) or 2-hydroxybenzaldehydes (**6**) (10 mmol), N-(R'-phenyl)chloroacetamides (**1-4**), chloroacetamide (**5**) 2-chloro-1-[5-(4-methoxyphenyl)-3-phenyl-4,5-dihydropyrazol-1-yl]-ethanone (**6**) (10 mmol) and potassium hydroxide (10 mmol) for 2 h in the anhydrous ethanol medium. Separate the resulting solution and distill in vacuum. Recrystallize the precipitate from ethanol or isopropanol.

2-(4-Chloro-2-formylphenoxy)-N-(2-trifluoromethylphenyl)-acetamide (**1**). Yield – 82%. M.p. – 154-157°C. ^{1}H NMR, δ, ppm, (*J*, Hz): 4.60s (2H, CH_2), 6.80 d (1H, C_6H_3, *J*=8.1Hz), 7.00 d (1H, C_6H_3, *J*=8.1 Hz), 7.05 d (1H, C_6H_3, *J*=2.7Hz), 7.10-7.30 m (4H, C_6H_4), 10.20s (1H, CHO), 13.90 br.s (1H, NH). Calculated, %: C53.90, H 3.30, N 4.00. $C_{16}H_{11}ClF_3NO_3$. Found, %: C 53.72, H 3.10, N 3.92.

2-(4-Chloro-2-formylphenoxy)-N-(4-fluorophenyl)-acetamide (**2**). Yield – 78%. M.p. – 169-171°C. ^{1}H NMR, δ, ppm, (*J*, Hz): 4.55 s (2H, CH_2), 6.90 d (1H, C_6H_3, *J*=8.0 Hz), 7.00 t (1H, C_6H_4, *J*=7.6 Hz),7.10 d (1H, C_6H_3, *J*=8.0 Hz), 7.20 d (1H, C_6H_3, *J*=2.9 Hz), 7.30 d (1H, C_6H_4, *J*=0.7Hz), 7.50 t (1H, C_6H_4, *J*=7.6 Hz), 7.60 d (1H, C_6H_4, *J*=0.7 Hz), 9.95 s (1H, CHO), 13.50 br.s (1H, NH). Calculated, %: C 58.70, H 3.70, N 4.60. $C_{15}H_{11}ClFNO_3$. Found, %: C 58.55, H 3.60, N 4.55.

2-(4-Chloro-2-formylphenoxy)-N-(4-methoxyphenyl)-acetamide (**3**). Yield – 84%. M.p. – 184-187°C. ^{1}H NMR, δ, ppm, (*J*, Hz): 3.55 s (3H, CH_3), 4.55s (2H, CH_2), 6.85 d (1H, C_6H_3, *J*=7.8 Hz), 6.95 d (1H, C_6H_3, *J*=7.8 Hz), 7.15d (1H, C_6H_3, *J*=2.7 Hz), 7.20 d (2H, C_6H_4, *J*=8.0 Hz), 7.40 d (2H, C_6H_4, *J*=8.0 Hz), 10.20 s (1H, CHO), 13.60 br.s (1H, NH). Calculated, %: C 60.00, H 4.70, N 4.55. $C_{16}H_{14}ClNO_4$. Found, %: C 60.10, H 4.41, N 4.38.

4-[2-(4-Chloro-2-formylphenoxy)-acetylamino]-benzoic acid ethyl ester (**4**). Yield – 81%. M.p. – 173-175°C.^{1}HNMR, δ, ppm, (*J*, Hz): 1.20 t (3H, CH_3), 4.10 q (2H, CH_2), 4.45 s (2H, CH_2), 6.90 d (1H, C_6H_3, *J*=8.0 Hz), 7.20 d (1H, C_6H_3, *J*=8.0 Hz), 7.30 d (1H, C_6H_3, *J*=2.9 Hz), 7.50 d (2H, C_6H_4, *J*=8.2 Hz), 7.70 d (2H, C_6H_4, *J*=8.2 Hz), 10.20 s (1H, CH), 11.50 br.s (1H, NH). Calculated, %: C 59.70, H 4.60, N 4.00. $C_{18}H_{16}ClNO_5$. Found, %: C 59.76, H 4.46, N 3.87.

2-(4-Chloro-2-formylphenoxy)acetamide (**5**). Yield – 75%. M.p. – 179-181°C. ^{1}H NMR, δ, ppm, (*J*, Hz): 4.40 s (2H, CH_2), 6.80 d (1H, C_6H_3, *J*=7.4 Hz), 7.00 d (1H, C_6H_3, *J*=2.3 Hz), 7.10 d (1H, C_6H_3, *J*=7.4 Hz), 7.20 s (2H, NH), 10.10s (1H, CHO), 13.60 br.s (1H, NH). Calculated, %: C 50.70, H 3.70, N 6.60. $C_9H_8ClNO_3$. Found, %: C 50.60, H 3.77, N 6.56.

2-{2-[5-(4-Methoxyphenyl)-3-phenyl-4,5-dihydro-pyrazol-1-yl]-2-oxoethoxy}-benzaldehyde (**6**). Yield – 84%. M.p. – 168-171°C.^{1}H NMR, δ, ppm, (*J*, Hz): 3.30 dd (1H, CH_2, *J*=18.0; 4.0 Hz), 3.75 s (3H, OCH_3), 4.00 dd (1H, CH_2, *J*=17.9;10.9Hz), 4.60s (2H, CH_2), 5.70 dd (1H, CH_2, *J*=12.6; 3.9 Hz), 6.80 t (1H, C_6H_4, *J*=8.3Hz), 6.85d (2H, C_6H_4, *J*=8.7 Hz), 7.15d (2H, C_6H_4, *J*=8.7 Hz), 7.25 d (1H, C_6H_4, *J*=0.7 Hz), 7.30–7.40 m (3H, C_6H_5), 7.65 t (1H, C_6H_4, *J*=8.3 Hz), 7.75 d (1H, C_6H_4, *J*=0.7 Hz), 7.85 d (2H, C_6H_4, *J*=8.7 Hz), 10.30s (1H, CHO). Calculated, %: C 72.70, H 5.70, N 6.80. $C_{25}H_{22}N_2O_4$. Found, %: C 72.45, H 5.35, N 6.76.

***The general procedure for the synthesis of ylidene derivatives* 7-9 *and* 11-13**. Reflux the mixture of the appropriate oxocompounds **1-3** or **5-6** (10 mmol), 2-thioxo-4-thiazolidinone (**7-9**, **11**, **13**) or 2-(4-hydroxyphenyl)imino-4-thiazolidinone (**12**) (10 mmol) and anhydrous sodium acetate (15mmol) for 2 h in a glacial acetic acid (5 ml). Filter the powders obtained, wash with ethanol and recrystallize with the corresponding solvent.

***The procedure for the synthesis of ylidene derivative* 10.** Reflux the mixture of 1,2,4-triazole-3(5)-thiol (10 mmol), chloroacetic acid (10 mmol), oxocompound **4** (10 mmol) and anhydrous sodium acetate (15 mmol) for 2 h in the mixture of acetic anhydride (5 ml) and glacial acetic acid (5 ml). Filter the powders obtained, wash with ethanol and recrystallize with acetic acid.

2-[4-Chloro-2-(4-oxo-2-thioxo-thiazolidin-5-ylidene)phenoxy]-N-(2-trifluoromethylphenyl)

acetamide (**7**)**.** Yield – 67%. M.p. – 236 (with decomp.) °C. ^{1}H NMR, δ, ppm, (*J*, Hz): 4.90 s (2H, CH_2), 7.10 d (1H, C_6H_3, *J*=8.2 Hz), 7.30 d (1H, C_6H_3, *J*=8.2 Hz), 7.35 s (1H, C_6H_3, *J*=2.9 Hz), 7.40-7.60 m (4H, C_6H_4), 7.90 s (1H, CH), 9.50 s (1H, NH), 13.60 br.s (1H, NH). Calculated, %: C 48.50, H 2.70, N 5.80. $C_{19}H_{12}ClF_3N_2O_3S_2$. Found, %: C 48.26, H 2.56, N 5.92.

2-[4-Chloro-2-(4-oxo-2-thioxo-thiazolidin-5-ylidene)phenoxy]-N-(4-chlorophenyl)acetamide (**8**)**.** Yield – 71%. M.p. – 242-244°C. ^{1}H NMR, δ, ppm, (*J*, Hz): 4.80 s (2H, CH_2), 7.10 d (1H, C_6H_3, *J*=7.8 Hz), 7.30 d (1H, C_6H_3, *J*=7.8 Hz), 7.35 d (1H, C_6H_3, *J*=2.6 Hz), 7.00 t (1H, C_6H_4, *J*=7.6 Hz), 7.40 d (1H, C_6H_4, *J*=0.5 Hz), 7.60 t (1H, C_6H_4, *J*=7.8 Hz), 7.70 d (1H, C_6H_4, *J*=0.5 Hz), 7.90 s (1H, CH),10.00 s (1H, NH). Calculated, %: C 51.30, H 2.80, N 6.70. $C_{18}H_{12}ClFN_2O_3S_2$. Found, %: C 51.12, H 2.86, N 6.62.

2-[4-Chloro-2-(4-oxo-2-thioxo-thiazolidin-5-ylidene)phenoxy]-N-(4-methoxyphenyl)acetamide (**9**)**.** Yield – 74%. M.p. – 248 (with decomp.) °C. ^{1}H NMR, δ, ppm, (*J*, Hz): 3.65 s (3H, OCH_3), 4.60 s (2H, CH_2), 7.10 d (1H, C_6H_3, *J*=7.5 Hz), 7.15 d (1H, C_6H_3, *J*=7.5 Hz), 7.20 d (1H, C_6H_3, *J*=2.4 Hz), 7.30 d (2H, C_6H_4, *J*=7.8 Hz), 7.50 d (2H, C_6H_4, *J*=7.8 Hz), 7.90 s (1H, CH), 10.00 s (1H, NH), 11.30 br.s (1H, NH). Calculated, %: C 52.60, H 3.60, N 6.60. $C_{19}H_{15}ClN_2O_4S_2$. Found, %: C 52.47, H 3.48, N 6.44.

4-{2-[4-Chloro-2-(6-oxothiazolo[3,2-b][1,2,4]triazol-5-ylidenemethyl)-phenoxy]-acetylamino}-benzoic acid ethyl ester (**10**)**.** Yield – 83%. M.p. – 176-178°C. ^{1}H NMR, δ, ppm, (*J*, Hz): 1.30 t (3H, CH_3), 4.30 q (2H, CH_2), 4.90 s (2H, CH_2), 7.10 d (1H, C_6H_3, *J*=7.3 Hz), 7.40 d (1H, C_6H_3, *J*=7.3 Hz), 7.50 d (1H, C_6H_3, *J*=2.2 Hz), 7.70 d (2H, C_6H_4, *J*=7.7 Hz), 7.90 d (2H, C_6H_4, *J*=7.7 Hz), 8.25 s (1H, CH), 8.50s (1H, CH), 10.40 s (1H, NH), 11.60 br.s (1H, NH). Calculated, %: C 54.50, H 3.50, N 11.70. $C_{22}H_{17}ClN_4O_5S$. Found, %: C 54.49, H 3.53, N 11.55.

2-[4-Chloro-2-(4-oxo-2-thioxo-thiazolidin-5-ylidene)phenoxy]-acetamide (**11**). Yield – 63%. M.p. – 240 (with decomp.) °C. ^{1}H NMR, δ, ppm, (*J*, Hz): 4.60s (2H, CH_2), 7.00 d (1H, C_6H_3, *J*=7.9 Hz), 7.40 d (1H, C_6H_3, *J*=7.9 Hz), 7.50 d (1H, C_6H_3, *J*=2.4 Hz), 7.60 s (2H, NH), 7.80s (1H, CH), 13.80 br.s (1H, NH). Calculated, %: C 43.70, H 2.70, N 8.60. $C_{12}H_9ClN_2O_3S_2$. Found, %: C 43.84, H 2.76, N 8.52.

2-{4-Chloro-2-[2-(4-hydroxyphenyl)imino-4-oxo-thiazolidin-5-yliden]-phenoxy}-N-(4-methoxyphenyl)acetamide (**12**)**.** Yield – 81%. M.p. > 250°C. ^{1}H NMR, δ, ppm, (*J*, Hz): 3.75 s (3H, CH_3), 4.75 s (2H, CH_2), 6.80 d (2H, C_6H_4, *J*=7.8 Hz), 6.90 d (2H, C_6H_4, *J*=7.8 Hz), 7.00 d (1H, C_6H_3, *J*=8.2 Hz), 7.10 d (1H, C_6H_3, *J*=8.2 Hz), 7.30 d (1H, C_6H_3, *J*=2.7 Hz), 7.40 d (2H, C_6H_4, *J*=8.1 Hz), 7.60 d (2H, C_6H_4, *J*=8.1 Hz), 8.00s (1H, CH), 9.40 s (1H, OH), 9.75s (1H, NH), 11.20 br.s (1H, NH). Calculated, %: C 59.00, H 3.90, N 8.50. $C_{25}H_{20}ClN_3O_5S$. Found, %: C 58.88, H 3.95, N 8.24.

5-(2-{2-[5-(4-Methoxyphenyl)-3-phenyl-4,5-dihydropyrazol-1-yl]-2-oxoethoxy}-benzeliden)-2-thioxo-thiazolidin-4-one (**13**). Yield – 83%. M.p. – 233-235°C. ^{1}H NMR, δ, ppm, (*J*, Hz): 3.40 dd (1H, CH_2, *J*=17.8, 3.9Hz), 3.80 s (3H, OCH_3), 4.15 dd (1H, CH_2, *J*=18.1, 10.7Hz), 4.70 s (2H, CH_2), 5.90 dd (1H, CH_2, *J*=12.4, 3.8Hz), 7.00 t (1H, C_6H_4, *J*=8.5Hz), 7.05 d (2H, C_6H_4, *J*=8.7 Hz), 7.20 d (2H, C_6H_4, *J*=8.7 Hz), 7.30 d (1H, C_6H_4, *J*=0.9 Hz), 7.35–7.50 m (3H, C_6H_5), 7.70 t (1H, C_6H_4, *J*=8.5 Hz), 7.80 d (1H, C_6H_4, *J*=0.9 Hz), 7.90 d (2H, C_6H_4, *J*=8.7 Hz), 8.50 s (1H, CH), 9.90 s (1H, NH). Calculated, %: C 63.60, H 4.50, N 7.90. $C_{28}H_{23}N_3O_3S_4$. Found, %: C 63.50, H 4.38, N 7.93.

Conclusions

1. The effective synthetic method for 2-(4-R-2-formylphenoxy)-N-(R'-phenyl)acetamides, which are suitable "building blocks" for the structural design of new potential bioactive 4-thiazolidinones has been developed.

2. A series of 5-ylidene-4-thiazolidinones from 2-(4-R-2-formylphenoxy)-N-(R'-phenyl)acetamides has been synthesized and their anti-exudative activity in the carrageenin paw edema test in mice has been studied.

3. It has been found that all compounds synthesized have a significant anti-inflammatory activity, and the "lead-compound" – 5-ylidene derivative of thiazolo[3,2-b][1,2,4]triazole-6-one exhibiting the anti-exudative activity equivalent to the classic NSAID Diclofenac with the low acute toxicity level has been identified.

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THE SYNTHESIS OF 6-R-2,2,4-TRIMETHYL-1,2-DIHYDROQUINOLINE- AND 6-R-4-R'-2,2,4-TRIMETHYL-1,2,3,4-TETRAHYDROQUINOLINE-8-CARBOXYLIC ACIDS – THE STRUCTURAL ANALOGUES OF HELQUINOLINE

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Key words: pyrrolo[3,2,1-ij]quinoline-1,2-diones; oxidation; 6-R-2,2,4-trimethyl-1,2-dihydroquinoline-8-carboxylic acid; 6-R-4-R'-2,2,4-trimethyl-1,2,3,4-tetrahydroquinoline-8-carboxylic acid

The peculiarities of the oxidation reaction of substituted (5,6-dihydro)-4,4,6-trimethyl-4H-pyrrolo[3,2,1-ij] quinoline-1,2-diones have been investigated. 6-R-2,2,4-trimethyl-1,2-dihydroquinoline-8-carboxylic acids and 6-R-4-R'-2,2,4-trimethyl-1,2,3,4-tetrahydroquinoline-8-carboxylic acids, which are structural analogues of the natural antibiotic Helquinoline ((2R,4S)-4-methoxy-2-methyl-1,2,3,4-tetrahydroquinoline-8-carboxylic acid), have been obtained by oxidation of 8-R-4,4,6-trimethyl-4H-pyrrolo[3,2,1-ij]quinoline-1,2-diones and their hydrogenated analogues – 8-R-6-R'-4,4,6-trimethyl-5,6-dihydro-4H-pyrrolo[3,2,1-ij]quinoline-1,2-diones. It has been shown that 8-R-6-R'-4,4,6-trimethyl-5,6-dihydro-4H-pyrrolo[3,2,1-ij]quinoline-1,2-diones and 8-R-4,4,6-trimethyl-4H-pyrrolo [3,2,1-ij]quinoline-1,2-diones are oxidized similar to isatin with opening of the pyrrole-1,2-dione fragment and subsequent decarboxylation, and the presence of bulky substituents – gem-dimethyl groups in the second position of the hydroquinoline cycle has no steric effect on the process. Moreover, it has been found that oxidation of 8-R-4,4,6-trimethyl-4H-pyrrolo[3,2,1-ij]quinoline-1,2-diones proceeds selectively with opening the pyrrole-1,2-dione fragment without affecting the multiple bond of the dihydroquinoline cycle, polymerization also does not occur on it. The structure of 6-R-4-R'-2,2,4-trimethyl-1,2,3,4-tetrahydroquinoline-8-carboxylic acids and 6-R-2,2,4-trimethyl-1,2-dihydroquinoline-8-carboxylic acids has been confirmed by ^{1}H NMR and ^{13}C NMR spectroscopy, mass spectrometry and elemental analysis. With the help of mass spectroscopy it has been shown that the heterocyclic fragment of 6-R-2,2,4-trimethyl-1,2-dihydroquinoline-8-carboxylic acids is more stable compared to the fragment of 6-R-4-R'-2 2,4-trimethyl-1,2,3,4-tetrahydroquinoline-8-carboxylic acids.

СИНТЕЗ 6-R-2,2,4-ТРИМЕТИЛ-1,2-ДИГІДРОХІНОЛІН- І 6-R-4-R'-2,2,4-ТРИМЕТИЛ-1,2,3,4-ТЕТРАГІДРОХІНОЛІН-8-КАРБОНОВИХ КИСЛОТ – СТРУКТУРНИХ АНАЛОГІВ HELQUINOLINE
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Ключові слова: *пірол[3,2,1-ij]хінолін-1,2-діон; окиснення; 6-R-2,2,4-триметил-1,2-дигідрохінолін-8-карбонова кислота; 6-R-4-R'-2,2,4-триметил-1,2,3,4-тетрагідрохінолін-8-карбонова кислота*
Досліджені особливості реакції окиснення в ряду заміщених (5,6-дигідро)-4,4,6-триметил-4Н-піроло[3,2,1-ij] хінолін-1,2-діонів. Окисненням 8-R-4,4,6-триметил-4Н-піроло[3,2,1-ij]хінолін-1,2-діонів і їх гідрованих аналогів 8-R-6-R'-4,4,6-триметил-5,6-дигідро-4Н-піроло[3,2,1-ij]хінолін-1,2-діонів отримані відповідно 6-R-2,2,4-триметил-1,2-дигідрохінолін-8-карбонові кислоти та 6-R-4-R'-2,2,4-триметил-1,2,3,4-тетрагідрохінолін-8-карбонові кислоти, що є структурними аналогами природного антибіотика Helquinoline ((2R, 4S)-4-метокси-2-триметил-1,2,3,4-тетрагідрохінолін-8-карбонової кислоти). Показано, що 8-R-6-R'-4,4,6-триметил-5,6-дигідро-4Н-піроло[3,2,1-ij]хінолін-1,2-діони і 8-R-4,4,6-триметил-4Н-піроло[3,2,1-ij]хінолін-1,2-діони окиснюються подібно ізатину з розкриттям пірол-1,2-діонового фрагменту і подальшим декарбоксилюванням, причому наявність у другій позиції гідрохінолінового циклу об'ємних заступників – гем-диметильних груп не чинить стеричного впливу на цей процес. Крім того, встановлено, що окиснення 8-R-4,4,6-триметил-4Н-піроло[3,2,1-ij]хінолін-1,2-діонів протікає селективно з розкриттям пірол-1,2-діонового фрагменту, не зачіпаючи кратну зв'язку дигідрохінолінового циклу, полімеризація по ній також не відбувається. Будову 6-R-4-R'-2,2,4-триметил-1,2,3,4-тетрагідрохінолін-8-карбонових кислот і 6-R-2,2,4-триметил-1,2-дигідрохінолін -8-карбонових кислот підтверджено даними ЯМР ^{1}H та ЯМР ^{13}C спектроскопії, мас-спектрометрії та елементного аналізу. За допомогою мас-спектроскопії показано, що більшою стабільністю володіє гетероциклічний фрагмент 6-R-2,2,4-триметил-1,2-дигідрохінолін-8-карбонових кислот у порівнянні з фрагментом 6-R-4-R'-2, 2,4-триметил-1,2,3,4-тетрагідрохінолін-8-карбонових кислот.

СИНТЕЗ 6-R-2,2,4-ТРИМЕТИЛ-1,2-ДИГИДРОХИНОЛИН- И 6-R-4-R'-2,2,4-ТРИМЕТИЛ-1,2,3,4-ТЕТРАГИДРОХИНОЛИН-8-КАРБОНОВЫХ КИСЛОТ – СТРУКТУРНЫХ АНАЛОГОВ HELQUINOLINE
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Ключевые слова: *пирроло[3,2,1-ij]хинолин-1,2-дионы; окисление; 6-R-2,2,4-триметил-1,2-дигидрохинолин-8-карбоновая кислота; 6-R-4-R'-2,2,4-триметил-1,2,3,4-тетрагидрохинолин-8-карбоновая кислота*
Изучены особенности реакции окисления в ряду замещенных (5,6-дигидро)-4,4,6-триметил-4Н-пирроло [3,2,1-ij]хинолин-1,2-дионов. Окислением 8-R-4,4,6-триметил-4Н-пирроло[3,2,1-ij]хинолин-1,2-дионов и их гидрированных аналогов 8-R-6-R'-4,4,6-триметил-5,6-дигидро-4Н-пирроло[3,2,1-ij]хинолин-1,2-дионов получены соответственно 6-R-2,2,4-триметил-1,2-дигидрохинолин-8-карбоновые кислоты и 6-R-4-R'-2,2,4-триметил-1,2,3,4-тетрагидрохинолин-8-карбоновые кислоты, являющиеся структурными ана-

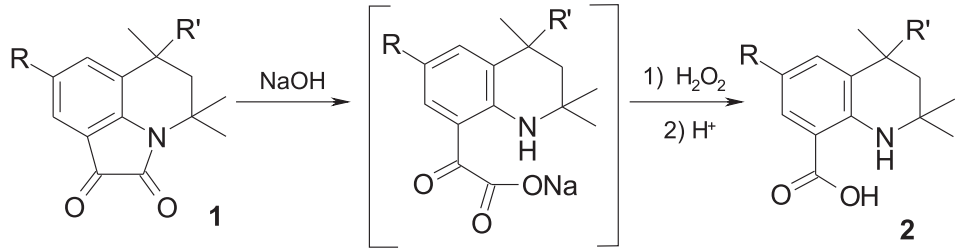
логами природного антибиотика Helquinoline ((2R,4S)-4-метокси-2-метил-1,2,3,4-тетрагидрохинолин-8-карбоновой кислоты). Показано, что 8-R-6-R'-4,4,6-триметил-5,6-дигидро-4H-пирроло[3,2,1-ij]хинолин-1,2-дионы и 8-R-4,4,6-триметил-4H-пирроло[3,2,1-ij]хинолин-1,2-дионы окисляются подобно изатину с раскрытием пиррол-1,2-дионового фрагмента и последующим декарбоксилированием, причем наличие во втором положении гидрохинолинового цикла объемных заместителей – гем-диметильных групп не оказывает стерического влияния на этот процесс. Кроме того, установлено, что окисление 8-R-4,4,6-триметил-4H-пирроло[3,2,1-ij]хинолин-1,2-дионов протекает селективно с раскрытием пиррол-1,2-дионового фрагмента, не затрагивая кратную связь дигидрохинолинового цикла, полимеризация по ней также не происходит. Строение 6-R-4-R'-2,2,4-триметил-1,2,3,4-тетрагидрохинолин-8-карбоновых кислот и 6-R-2,2,4-триметил-1,2-дигидрохинолин-8-карбоновых кислот подтверждено данными ЯМР ^{1}H и ЯМР ^{13}C спектроскопии, масс-спектрометрии и элементного анализа. С помощью масс-спектроскопии показано, что большей стабильностью обладает гетероциклический фрагмент 6-R-2,2,4-триметил-1,2-дигидрохинолин-8-карбоновых кислот по сравнению с фрагментом 6-R-4-R'-2,2,4-триметил-1,2,3,4-тетрагидрохинолин-8-карбоновых кислот.

The fragment of tetrahydroquinoline carboxylic acid is the structural moiety of a wide range of natural quinoline alkaloids such martinellic acid isolated from the root bark of the South American plant *Martinella iquitosensis* and a new natural antibiotic *Helquinoline* ((2R, 4S) -4-methoxy-2-methyl-1,2, 3,4-tetrahydro-8-quinolinecarboxylic acid) obtained from *Janibacter limosus Hel* [1-5]. It is also present in the structure of some synthetic medicinal products (antibiotic of a new generation virantmycin [6, 7], oxamniquine [8] used to treat schistosomiasis, etc.). In particular, hydroquinoline-8-carboxylic acids possess the antirheumatic, antibacterial activity [4, 9]. At the same time derivatives of 2,2,4-trimethyl-1,2-dihydroquinolines and their hydrogenated analogues – substituted 2,2,4-trimethyl-1,2,3,4-tetrahydroquinolines [10-12] exhibit a broad spectrum of the biological activity. In this connection the synthesis of 2,2,4-trimethyl-1,2-dihydroquinoline and 2,2,4-trimethyl-1,2, 3,4-tetrahydroquinoline-8-carboxylic acids being the structural analogues of the natural antibiotic *Helquinoline* is of interest. In addition, these aromatic amino acids are good building blocks for constructing new heterocyclic compounds, including with the properties of surfactants.

One of the most effective methods for the synthesis of substituted anthranilic acids is oxidation of various 1*H*-indole-2,3-dione (isatins) with hydrogen peroxide in the aqueous solution of alkalies [13, 14]. In the cause of this reaction isatins undergo disclosure of the pyrrole ring and are converted into salts of isatoic acid decarboxylated when reacting with an oxidizing agent. The structure of various 4,4,6-trimethyl-4*H*-pyrrolo[3,2,1-*ij*]quinoline-1,2-diones [15] earlier synthesized by us and the structure of isatin contain the pyrrol-1,2-dione fragment, which can be subjected to oxidation, but the presence of *gem*-dimethyl groups may create steric hindrance for the attack of the carbon atom of the amide group by the hydroxide ion [16]. It has been found that like isatin the disclosure of the pyrrole-1,2-dione fragment and decarboxylation occur without difficulty for all 8-R-6-R'-4,4,6-trimethyl-5,6-dihydro-4*H*-pyrrolo[3,2,1-*ij*] quinolin-1,2-diones **1a-f** under the action of hydrogen peroxide in the alkaline medium. The subsequent treatment with dilute hydrochloric acid leads to formation of 6-R-4-R'-2,2,4-trimethyl-1,2,3,4-tetrahydroquinoline-8-carboxylic acids **2a-f** undescribed previously (Scheme 1).

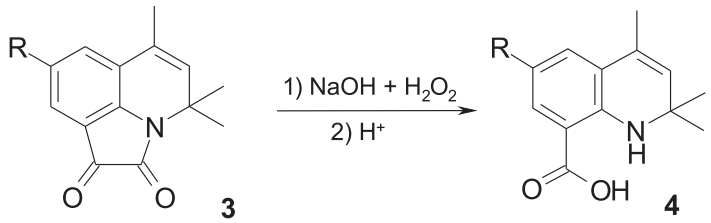
Similar interaction was conducted with hydrogen peroxide in the alkaline solution of 8-R-4,4,6-trimethyl-4*H*-pyrrolo[3,2,1-ij]quinoline-1,2-diones **3a-e**, containing the multiple bond in the heterocycle. It should be noted that for 1.2-dihydroquinolines polymerization is possible [17]. Furthermore, it is known that *N*-acyl-1,2-dihydroquinoline are oxidized with peroxides to form *N*-acyl-1,2-dihydroquinoline-3,4-epoxides [18], in which opening of the epoxide ring occurs under the action of bases [19]. It has been found that the oxidation reaction of pyrroloquinolinediones **3a-e** occurs selectively without affecting the multiple bond, and leads to 6-R-2,2,4-trimethyl-1,2-dihydroquinoline-8-carboxylic acids **4a-e**. Formation of polymerization products in this reaction is not detected (Scheme 2).

The structure of quinolinecarboxylic acid **2a-f** and **4a-e** has been unequivocally proven by the totality of the evidence of spectroscopy and spectrometry. The signals of protons of the secondary amino- and carboxyl groups are present in the ^{1}H NMR spectra of compounds **2a-f** and **4a-e** compared to the spec-



1, **2** R = H (a), Me (b, f), MeO (c), Et (d), F (e); R' = H (a-e), Ph (f)

Scheme 1



3, **4** R = H (a), Me (b), MeO (c), Et (d), F (e)

Scheme 2

tra of the starting pyrroloquinolinediones **1a-f** and **3a-e**. In the mass spectra (EI) of quinolinecarboxylic acids **2a-f** and **4a-e** the peaks of molecular ions with the low intensity (I_{rel} = 20-25%, I_{rel} = 10-17%, respectively) are observed; they are subjected to further fragmentation with emission of the methyl radical. The ions formed by sequential cleaving of molecular ions of the methyl radical and the molecule of H_2O have the maximum intensity (I_{rel} = 100%). In the spectra of 1,2-dihydroquinoline-8-carboxylic acids **4a-e** in contrast to the spectra 1,2,3,4-tetrahydroquinoline-8-carboxylic acids **2a-f** the fragment ions formed by sequential cleaving of molecular ions of the methyl radical, the molecule of H_2O and the molecule of CO (I_{rel} = 15-20%) are present. It indicates a greater stability of the 1,2-dihydroquinoline fragment compared to the 1,2,3,4-tetrahydroquinoline one.

Experimental Part

The ^{1}H NMR spectra and ^{13}C were recorded on a "Bruker AM-500" device (500 and 125 MHz, respectively) in the pulsed Fourier regime in $CDCl_3$, the position of signals of the test substances was determined by the δ-scale. The assignment of signals was carried out relative to the residual proton signals of the deuterium solvent. Mass spectra were recorded on a FINNIGAN MAT.INCOS spectrometer with the electron impact of 50 70 evc and direct input of the sample into the source of ions at 100-150°C and an accelerating voltage of 70 eV. Elemental analysis was performed on a Perkin Elmer 2400 device. The melting points were determined on a PTP-M device. Control of the individual reactants and the compounds obtained, as well as the course of the reaction was monitored by thin layer chromatography (TLC) on Merck TLC Silica gel 60 F254 plates (254-subscript) in the system of $CHCl_3$-EtOAc, 10:1. The starting compounds **1a-f** and 3**a-e** were synthesized by the procedure given earlier [15].

The general procedure for oxidation of 8-R-6-R'-4,4,6-trimethyl-5,6-dihydro-4H-pyrrolo[3,2,1-ij] quinoline-1,2-diones 1a-f and 8-R-4,4,6-trimethyl-4H-pyrrolo[3,2,1-ij]quinoline-1,2-diones 3a-e. Allow to stand 0.02 Mol of the corresponding pyrroloquinolinedione **1a-f** or **3a-e** in 20 ml of 20% aqueous solution of sodium hydroxide for 30 min, then while stirring and cooling add 0.04 Mol of hydrogen peroxide and mix at room temperature for 1.5-2 h. Pour the reaction mixture into 200 ml of water, neutralize with 10% HCl, filter the precipitate formed, wash with water, dry, crystallize from CCl_4 to give slightly yellow powders **2a-f** and **4a-e**.

2,2,4-Trimethyl-1,2,3,4-tetrahydroquinoline-8-carboxylic acid 2a. Yield – 2.0 g, 89%. M.p. – 167-168°C. ^{1}H NMR spectrum, δ, ppm: 1.27 (s, 3H, CH_3); 1.35 (s, 6H, C $(CH_3)_2$); 1.37 (d, J = 6.5 Hz, 1H, CH_2); 1.76 - 1.80 (m, 1H, CH_2); 2.91-2.96 (m, 1H, CH); 6.58 (t, J = 7.5 Hz, 1H, H-6 Ar); 7.33 (d, J = 7.5 Hz, 1H, H-5 Ar); 7.83 (d, J = 7.5 Hz, 1H, H-7, Ar); 9.50-10.20 (br.s., 1H, NH); 11.20-12.50 (br.s., 1H, OH). 13C NMR spectrum, δ, ppm: 20.0, 27.5, 29.2, 31.5, 43.1, 49.4, 107.2, 113.7, 126.9, 130.2, 132.0, 148.0, 174.2. MS: m/e (%) 219 ([M$^+$] (19)), 204 ([M-CH_3] (50)), 186 ([M-CH_3-H_2O] (100)). Found, %: C 71.31; H 7.79; N 6.47. $C_{13}H_{17}NO_2$. Calculated, %: C 71.21; H 7.81; N 6.39.

2,2,4,6-Tetramethyl-1,2,3,4-tetrahydroquinoline-8-carboxylic acid 2b. Yield – 2.0 g, 82%. M.p. – 196-197°C. ^{1}H NMR spectrum, δ, ppm: 1.29 (s, 3H, CH_3); 1.35 (s, 6H, C $(CH_3)_2$); 1.37 (d, J = 6.8 Hz, 1H, CH_2); 1.80-1.84 (m, 1H, CH_2); 2.26 (s, 3H, 6-CH_3); 2.95-2.99 (m, 1H, CH); 7.20 (s, 1H, H-5 Ar); 7.26 (s, 1H, H-7, Ar); 9.80-10.40 (br.s., 1H, NH); 11.30-12.50 (br.s., 1H, OH). ^{13}C NMR spectrum, δ, ppm: 20.3, 27.5, 29.0, 31.4, 43.3, 49.4, 107.6, 123.2, 127.4, 129.5, 133.7, 145.6, 174.1. MS: m/e (%) 233 ([M$^+$] (24)), 218 ([M-CH_3] (50)), 200 ([M-CH_3-H_2O] (100)). Found, %: C 72.15; H 8.29; N 6.11. $C_{14}H_{19}NO_2$. Calculated, %: C 72.07; H 8.21; N 6.00.

2,2,4-Trimethyl-6-methoxy-1,2,3,4-tetrahydroquinoline-8-carboxylic acid 2c. Yield – 2.4 g, 92%. M.p. – 155-156°C. ^{1}H NMR spectrum, δ, ppm: 1.31 (s, 3H, CH3); 1.36 (s, 6H, C (CH3)2); 1.37 (d, J = 6.1 Hz, 1H, CH_2); 1.82-1.86 (m, 1H, CH_2);); 2.98-3.01 (m, 1H, CH); 3.78 (c, 3H, OCH_3); 7.00 (c, 1H, H-5 Ar); 7.34 (c, 1H, H-7, Ar); 9.40-10.20 (br.s., 1H, NH); 11.40-12.60 (br.s., 1H, OH). ^{13}C NMR spectrum, δ, ppm: 20.2, 28.2, 29.4, 31.7, 43.1, 49.5, 55.8, 107.3, 111.0, 123.5, 129.3, 143.0, 149.1, 173.7. MS: m/e (%) 249 ([M$^+$] (25)), 234 ([M-CH_3] (40)), 216 ([M-CH_3-H_2O] (100)). Found, %: C 67.61; H 7.57; N 5.54. $C_{14}H_{19}NO_3$. Calculated, %: C 67.45; H 7.68; N 5.62.

2,2,4-Trimethyl-6-ethyl-1,2,3,4-tetrahydroquinoline-8-carboxylic acid 2d. Yield – 2.3 g, 88%. M.p. – 157-158°C. ^{1}H NMR Spectrum, δ, ppm: 1.21 (t, J = 7.6 Hz, 3H, CH_3CH_2); 1.27 (s, 3H, CH_3); 1.34 (s, 6H, C $(CH_3)_2$); 1.37 (d, J = 6.6 Hz, 1H, CH_2); 1.77-

1.81 (m, 1H, CH_2); 2.54 (q, J = 7.6 Hz, 2H, CH_3CH_2); 2.93-2.96 (m, 1H, CH); 7.21 (s, 1H, H-5 Ar); 7.66 (s, 1H, H-7, Ar); 9.40-10.50 (br.s., 1H, NH); 11.10-12.30 (br.s., 1H, OH). ^{13}C NMR spectrum, δ, ppm: 15.8, 20.4, 27.6, 27.8, 29.1, 31.5, 43.3, 49.4, 107.2, 127.2, 128.3, 129.5, 132.5, 146.3, 174.1. MS: m/e (%) 247 ([M^+] (27)), 232 ([M-CH_3] (54)), 214 ([M-CH_3-H_2O] (100)). Found, %: C 72.98; H 8.44; N 5.75. $C_{15}H_{21}NO_2$. Calculated,%: C 72.84; H 8.56; N 5.66.

2,2,4-Trimethyl-6-fluoro-1,2,3,4-tetrahydroquinoline-8-carboxylic acid 2e. Yield – 2.0 g, 91%. M.p. – 172-173°C. ^{1}H NMR Spectrum, δ, ppm: 1.32 (c, 3H, CH_3); 1.37 (s, 6H, C $(CH_3)_2$); 1.38 (d, J = 6.2 Hz, 1H, CH_2); 1.86-1.90 (m, 1H, CH_2);); 2.99-3.02 (m, 1H, CH); 7.15 (s, 1H, H-5 Ar); 7.53 (s, 1H, H-7, Ar); 9.60-10.20 (br.s, 1H, NH); 11.40-12.70 (br.s, 1H, OH). ^{13}C NMR Spectrum, δ, ppm: 20.4, 28.4, 29.2, 31.7, 43.21, 50.8, 115.1, 116.5, 120.9, 122.2, 131.0, 147.1, 153.2, 173.2. MS: m/e (%) 237 ([M^+] (21)), 222 ([M - CH_3] (49)), 204 ([M-CH_3-H_2O] (100)). Found, %: C 77.81; H 7.48; N 4.63. $C_{14}H_{19}NO_3$. Calculated, %: C 77.65; H 7.49; N 4.53.

2,2,4,6-Tetramethyl-4-phenyl-1,2,3,4-tetrahydroquinoline-8-carboxylic acid 2f. Yield – 2.4 g, 77%. M.p. – 205-206°C. ^{1}H NMR Spectrum, δ, ppm: 0.89 (s, 3H, C $(CH_3)_2$); 1.35 (s, 3H, C$(CH_3)_2$); 1.77 (s, 3H, CHs); 2.06-2.12 (m, 1H, CH_2); 2.24 (s, 3H, 6-CH_3); 2.38-2.43 (m, 1H, CH_2); 7.18 (s, 1H, H-5 Ar); 7.14-7.27 (m, 5H, Ph); 7.79 (s, 1H, H-7, Ar); 8.40-10.00 (br.s., 2H, NH, OH). ^{13}C NMR Spectrum, δ, ppm: 20.2, 27.7, 28.9, 31.3, 43.3, 49.5, 107.2, 123.4, 126.2, 127.6, 129.0, 129.5, 129.7, 133.7, 137.4, 145.0, 174.3. MS: m/e (%) 309 ([M^+] (22)), 294 ([M - CH_3] (38)), 276 ([M-CH_3-H_2O] (100)). Found, %: C 77.72; H 7.37; N 4.64. $C_{20}H_{23}NO_2$. Calculated,%: C 77.64; H 7.49; N 4.53.

2,2,4-Trimethyl-1,2-dihydroquinoline-8-carboxylic acid 4a. Yield – 1.9 g, 86%. M.p. – 174-175°C. ^{1}H NMR Spectrum, δ, ppm: 1.36 (s, 6H, C $(CH_3)_2$); 1.98 (s, 3H, CH_3); 5.38 (s, 1H, CH); 6.53 (t, J = 7.8 Hz, 1H, H-6 Ar); 7.19 (d, J = 7.8 Hz, 1H, H-5 Ar); 7.78 (d, J = 7.8 Hz, 1H, H-7, Ar); 8.90-9.20 (br.s, 1H, NH); 10.80-11.80 (br.s, 1H, OH). ^{13}C NMR Spectrum, δ, ppm: 19.0, 32.3, 52.0, 107.2, 114.2, 122.2, 127.4, 128.5, 128.9, 131.3, 131.4, 147.7, 174.0. MS: m/e (%) 217 ([M^+] (10)), 202 ([M-CH_3] (47)), 184 ([M-CH_3-H_2O] (100)), 156 ([M-CH_3-H_2O-CO] (15)). Found, %: C 72.01; H 7.06; N 6.40. $C_{13}H_{15}NO_2$. Calculated, %: C 71.87; H 6.96; N 6.45.

2,2,4,6-Tetramethyl-1,2-dihydroquinoline-8-carboxylic acid 4b. Yield – 2.0 g, 81%. M.p. – 197-198°C. ^{1}H NMR Spectrum, δ, ppm: 1.35 (s, 6H, C $(CH_3)_2$); 1.98 (s, 3H, CH_3); 2.22 (s, 3H, 6-CH_3); 5.39 (s, 1H, CH); 7.03 (s, 1H, H-5 Ar); 7.59 (s, 1H, H-7, Ar); 8.80-9.40 (br.s, 1H, NH); 10.50-11.20 (br.s, 1H, OH). ^{13}C NMR Spectrum, δ, ppm: 19.0, 20.4, 32.0, 51.9, 107.0, 122.4, 123.2, 127.5, 128.9, 130.3, 130.6, 130.7, 147.7, 174.0. MS: m/e (%) 231 ([M^+] (17)), 216 ([M-CH_3] (50)), 198 ([M-CH_3-H_2O] (100)), 170 ([M-CH_3-H_2O-CO] (21)). Found, %: C 72.82; H 7.52; N 6.13. C14H17NO2. Calculated, %: C 72.70; H 7.41; N 6.06.

2,2,4-Trimethyl-6-methoxy-1,2-dihydroquinoline-8-carboxylic acid 4c. Yield – 2.2 g, 87%. M.p. – 172-173°C. ^{1}H NMR Spectrum, δ, ppm: 1.39 (s, 6H, C $(CH_3)_2$); 2.00 (s, 3H, CH_3); 3.79 (s, 3H, OCH_3); 5.51 (s, 1H, CH); 6.93 (s, 1H, H-5 Ar); 7.30 (s, 1H, H-7, Ar); 9.00-9.20 (br.s, 1H, NH); 10.60-11.90 (br.s, 1H, OH). ^{13}C NMR Spectrum, δ, ppm: 19.0, 31.6, 32.3, 52.2, 55.9, 107.3, 111.4, 119.4, 124.6, 127.4, 130.3, 142.6, 149.5, 173.5. MS: m/e (%) 247 ([M^+] (14)), 232 ([M-CH_3] (58)), 214 ([M-CH_3-H_2O] (100)), 186 ([M-CH_3-H_2O-CO] (7)). Found, %: C 68.11; H 7.07; N 5.53. $C_{14}H_{17}NO_3$. Calculated, %: C 68.00; H 6.93; N 5.66.

2,2,4-Trimethyl-6-ethyl-1,2-dihydroquinoline-8-carboxylic acid 4d. Yield – 2.1 g, 84%. M.p. – 178-179°C. ^{1}H NMR Spectrum, δ, ppm: 1.21 (t, J = 7.6 Hz, 3H, CH_3CH_2); 1.37 (s, 6H, C $(CH_3)_2$); 2.01 (s, 3H, CH_3); 2.53 (q, J = 7.6 Hz, 2H, CH_3CH_2); 5.41 (s, 1H, CH); 7.08 (s, 1H, H-5 Ar); 7.52 (s, 1H, H-7, Ar); 8.00-8.90 (br.s, 1H, NH); 10.70-11.40 (br.s, 1H, OH). ^{13}C NMR Spectrum, δ, ppm: 15.8, 19.1, 28.0, 32.2, 52.1, 107.3, 122.6, 127.6, 128.8, 129.3, 129.5, 129.8, 130.2, 145.6, 173.9. MS: m/e (%) 245 ([M^+] (13)), 230 ([M-CH_3] (68)), 2128 ([M-CH_3-H_2O] (100)), 184 ([M-CH_3-H_2O-CO] (15)). Found, %: C 73.53; H 7.74; N 5.84. $C_{15}H_{19}NO_2$. Calculated, %: C 73.44; H 7.81; N 5.71.

2,2,4-Trimethyl-6-fluoro-1,2-dihydroquinoline-8-carboxylic acid 4e. Yield – 1.9 g, 78%. M.p. – 169-170°C. ^{1}H NMR Spectrum, δ, ppm: 1.38 (s, 6H, C $(CH_3)_2$); 1.97 (s, 3H, CH_3); 5.48 (s, 1H, CH); 6.96 (s, 1H, H-5 Ar); 7.44 (s, 1H, H-7, Ar); 8.70-9.00 (br.s, 1H, NH); 10.20-11.60 (br.s, 1H, OH). ^{13}C NMR Spectrum, δ, ppm: 19.1, 32.5, 52.1, 107.0, 114.1, 122.4, 127.2, 114.1, 129.0, 131.5, 147.6, 161.4, 174.5. MS: m/e (%) 235 ([M^+] (19)), 220 ([M-CH_3] (48)), 202 ([M-CH_3-H_2O] (100)), 174 ([M-CH_3-H_2O-CO] (23)). Found, %: C 72.82; H 7.52; N 6.13. $C_{13}H_{14}FNO_2$. Calculated, %: C 66.37; H 6.00; N 5.95.

Conclusions

1. It has been shown that 8-R-6-R'-4,4,6-trimethyl-5,6-dihydro-4*H*-pyrrolo[3,2,1-*ij*]quinoline-1,2-diones and 8-R-4,4,6-trimethyl-4*H*-pyrrolo[3,2,1-*ij*] quinoline-1,2-diones are oxidized with hydrogen peroxide in the presence of alkali to form 6-R-4-R'-2,2 4-trimethyl-1,2,3,4-tetrahydroquinoline-8-carboxylic acids and 6-R-2,2,4-trimethyl-1,2-dihydroquinoline-8-carboxylic acids, respectively, and the presence of bulky substituents – gem-dimethyl groups in the second position of the hydroquinoline cycle has no steric effect on disclosure of the pyrrole-1,2-dione fragment and the subsequent decarboxylation.

2. It has been found that oxidation of 8-R-4,4,6-trimethyl-4H-pyrrolo[3,2,1-ij]quinoline-1,2-diones proceeds selectively with opening of the pyrrole-1,2-dione fragment without affecting the multiple bond of the dihydroquinoline cycle.

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N-ACYLATION OF AMINO-9,10-ANTHRAQUINONES BY THE SYSTEM OF STRONG CARBOXYLIC ACID – AMMONIUM THIOCYANATE

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Key words: amino-9,10-anthraquinones; carboxylic acids; ammonium thiocyanate; ammonium acetate; acylation

The significance of the acylation reaction of amines is presented in the literary reference information. The products of the reaction of the corresponding amides are important intermediates in obtaining practically useful compounds. It has been shown that the most common methods of acylfunctionalization of amines are acetylation, trifluoroacetylation and formylation, usually acid anhydrides or chlorides are used as acylating reagents in these reactions in the presence of highly toxic and expensive catalysts. The authors have developed an approach to the synthesis of a number of N-acylated amino-9,10-anthraquinones, which is based on the use of a new acylation system consisting of a strong organic acid and ammonium thiocyanate. It has been determined that 1-amino-9,10-anthraquinone and its derivatives in the presence of the two-fold excess of ammonium thiocyanate can be acetylated only by formic and trifluoroacetic acids. 2-Amino-9,10-anthraquinone additionally can be acetylated by mercaptoacetic and acetic acids. The scheme of the reaction discovered has been proposed, it involves in situ generation of ammonium acetate from carboxylic acid and ammonium thiocyanate, which serves as the acylating reagent.

***N*-АЦИЛЮВАННЯ АМІНО-9,10-АНТРАХІНОНІВ СИСТЕМОЮ СИЛЬНА КАРБОНОВА КИСЛОТА – ТІОЦІАНАТ АМОНІЮ**

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Ключові слова: *аміно-9,10-антрахінони; карбонові кислоти; тіоціанат амонію; ацетат амонію; ацилювання*

Наведена інформативна літературна довідка синтетичної значимості реакції ацилювання амінів, продукти якої відповідні аміди є важливими інтермедіатами при отриманні значного масиву практично корисних сполук. На основі аналізу літературних джерел виявлено, що в процесах ацилфункціоналізації амінів найпоширенішими є методи ацетилювання, трифтороацетилювання та формілювання, в яких, як правило, в ролі ацилюючих реагентів використовуються ангідриди або хлороангідриди кислот у присутності високотоксичних і дорогих каталізаторів. Авторами розроблено підхід до синтезу низки N-ацильованих аміно-9,10-антрахінонів, який грунтується на застосуванні нової ацилюючої системи сильна органічна кислота – тіоціанат амонію. На прикладах взаємодії аміно-9,10-антрахінонів із форміатною, ацетатною, тіоацетатною та трифтороацетатною кислотами в присутності двократного надлишку тіоціанату амонію з'ясовано вплив структури аміносубстрату та карбонової кислоти на перебіг реакції ацилювання і утворення антрахіноліламідів. Встановлено, що 1-аміно-9,10-антрахінон та його заміщені аналоги в присутності тіоціанату амонію схильні до ацилювання тільки форміатною та трифтороацетатною кислотами, натомість 2-аміно-9,10-антрахінон окрім форміатної та трифтороацетатної кислот утворює аміди під дією ацетатної та тіоацетатної кислот. Запропонована схема знайденої реакції, яка передбачає in situ генерування із карбонової кислоти та тіоціанату амонію ацетату амонію, який власне і виконує роль ацилюючого реагента.

***N*-АЦИЛИРОВАНИЕ АМИНО-9,10-АНТРАХИНОНОВ СИСТЕМОЙ СИЛЬНАЯ КАРБОНОВАЯ КИСЛОТА – ТИОЦИАНАТ АММОНИЯ**

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Ключевые слова: *амино-9,10-антрахиноны; карбоновые кислоты; тиоцианат аммония; ацилирование*

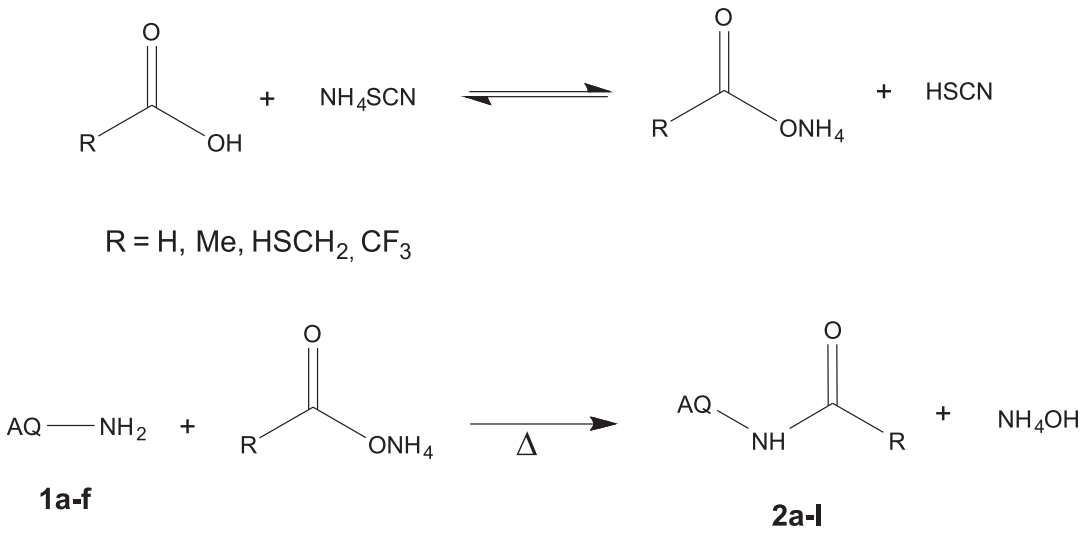
Приведена информативная литературная справка синтетической значимости реакции ацилирования аминов, продукты которой соответствующие амиды являются важными интермедиатами при получении значительного массива практически полезных соединений. На основе анализа литературных источников установлено, что в процессах ацилфункционализации аминов наиболее распространены методы ацетилирования, трифтор- ацетилирования и формилирования, в которых, как правило, в роли ацилирующих реагентов используются ангидриды или хлорангидриды кислот в присутствии высокотоксичных и дорогих катализаторов. Авторами разработан подход к синтезу ряда N-ацилированных амино-9,10-антрахинонов, основанный на применении новой ацилирующей системы сильная органическая кислота – тиоцианат аммония. На примерах взаимодействия амино-9,10-антрахинонов с муравьиной, уксусной, тиоуксусной и трифторуксусной кислотами в присутствии двухкратного избытка тиоцианата аммония выяснено влияние структуры аминосубстратов и карбоновой кислоты на протекание реакции ацилирования и образования антрахинолиламидов. Установлено, что 1-амино-9,10-антрахинон и его замещенные аналоги в присутствии тиоцианата аммония подвержены ацилированию только муравьиной и трифторуксусной кислотами, в то время как 2-амино-9,10-антрахинон кроме муравьиной и трифторуксусной кислот образует амиды под действием уксусной и тиоуксусной кислот. Предложена схема найденной реакции, которая предусматривает in situ генерирование с карбоновой кислоты и тиоцианата аммония ацетата аммония, который собственно и выполняет роль ацилирующего реагента.

Acylation of amines is one of the most common methods of their structural modifications, and it is widely used in organic synthesis and medicinal chemistry. *N*-Acyl residues are important protective groups, and the corresponding amides are effective intermediates in various chemical transformations aimed to obtain practically useful compounds. In the process of acylfunctionalization of amines the most generally used methods are: acetylation [1], trifluoroacetylation [2] and formylation [3-5]. The commonly used acylating reagents are acetic acid anhydride and chloride in the presence of highly toxic and expensive catalysts [6-9], trifluoroacetic acid anhydride and other highly electrophilic derivatives of trifluoroacetic acid [10-13], complexes of formic acid with carbodiimides [14, 15] or Lewis acids [16]. Thus, the search for environmentally benign and technologically convenient methods of acylation of amines by carboxylic acids with catalytic addition of cheap reagents is a topic of great interest [17-19].

N-Acylamino-9,10-anthraquinones became the subject of increased attention of researchers in recent years because of identification of 1-acetamide-9,10-anthraquinone as a new mutagenetic metabolite of 1-aminoanthracene [20]. 2-Trifluoroacetamide-9,10-anthraquinone was used as a selective colorimetric sensor for a cyanide anion in aqueous solutions [21]. The synthesis of *N*-acylamino-9,10-anthraquinones was carried out via the reaction with acetic [20, 22, 23], trifluoroacetic [21] acid anhydrides, and acetyl chloride [24].

We have shown that for this purpose a new acylating system consisting of a strong carboxylic acid and ammonium thiocyanate could be successfully used. It was found that the structure of aminosubstrate and carboxylic acid affected the acylation reactions of *N*-acylamino-9,10-anthraquinones **2a-l** on examples of reactions of 1- and 2-amino-9,10-anthraquinones (AQ-NH_2) **1a-f** with formic, acetic, mercaptoacetic, and trifluoroacetic acids in the presence of the two-fold excess of ammonium thiocyanate (Table). It was determined that 1-amino-9,10-anthraquinone **1a** and its derivatives **1b-d** were acylated only by formic and trifluoroacetic acids in the presence of ammonium thiocyanate. In the case of diamino-9,10-anthraquinones **1c,d** both amino groups took part in the reaction. 2-Amino-9,10-anthraquinone **1e** reacted not only with strong formic and trifluoroacetic acids, but it also gave amides with mercaptoacetic and acetic acids. On the contrary, 2-amino-3-chloro-9,10-anthraquinone **1f** underwent only trifluoroacylation, and isomeric 1-amino-2-chloro-9,10-anthraquinone was not acylated by any of the acids tested.

The regularities found well correlate with the electronic parameters of amino-9,10-anthraquinones, as well as with acidity of carboxylic acids. Thus, less basic 1-amino-9,10-anthraquinones **1a-d** gave the corresponding amides **2a-g** only with relatively strong trifluoroacetic (pKa = 0.23) and formic (pKa = 3.73) acids. At the same time more basic 2-amino-9,10-anthraquinone **1e** gave amides not only with such strong acids as trifluoroacetic and formic acids, but with weaker mercaptoacetic (pKa = 3.83) and acetic (pKa = 4.76) acids. However, acylation did not proceed with propanoic or butanoic acids. The result of the reaction is quite unexpected because the system of inorganic (organic) acid and ammonium thiocyanate is normally used to generate *in situ* thiocyanic acid, which is a thiocarbamoyl reagent for weak bases [25]. Therefore, in the case of amino-9,10-anthraquinones **1** formation of antraquinoylthioureas was expected. In fact, an alternative reaction – acylation of amino-9,10-anthraquinones by ammonium carboxylate resulted from the reaction of ammonium thiocyanate with strong organic acids took place (Scheme). These results are consistent with the data published



Scheme

Table

Products of N-acylation of amino-9,10-anthraquinones **1a-f** by the system of strong carboxylic acid – ammonium thiocyanate

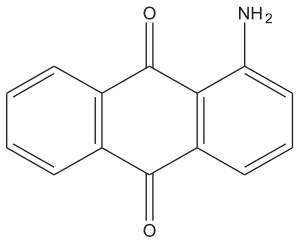
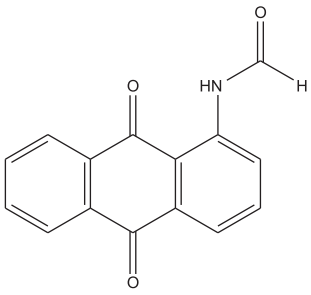
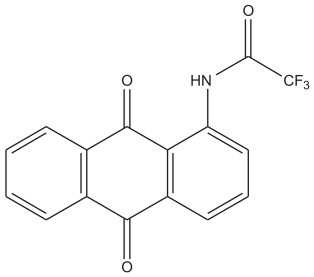
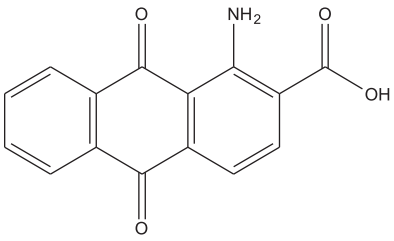
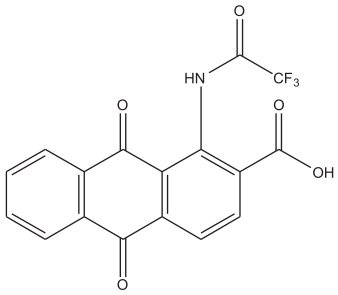
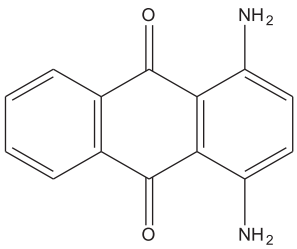
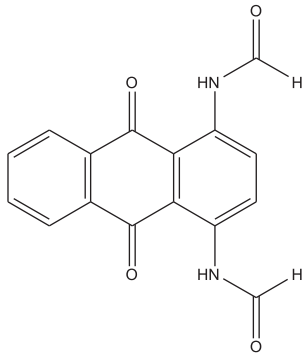
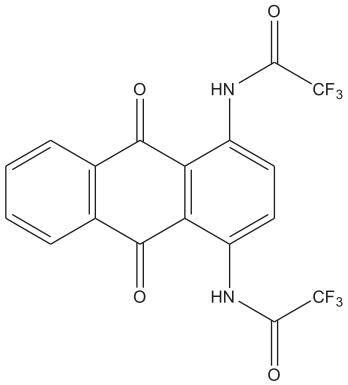
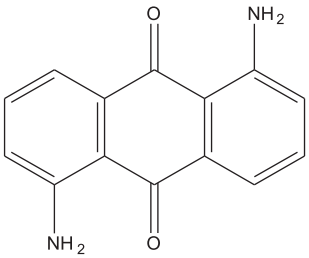
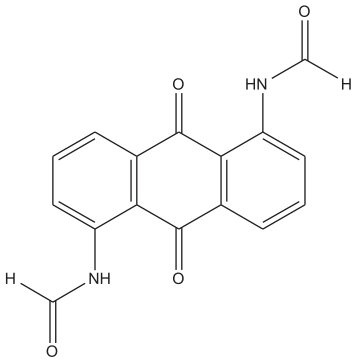
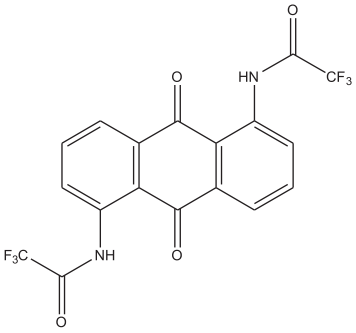
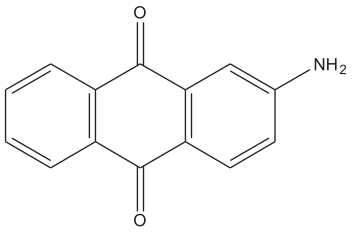
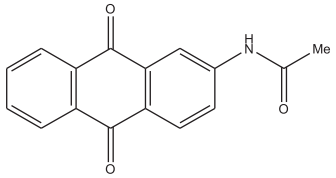
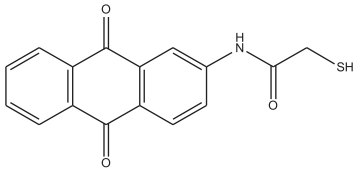
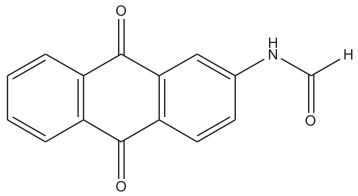
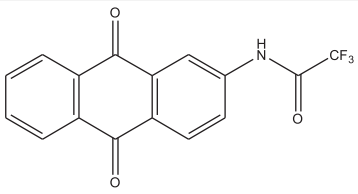
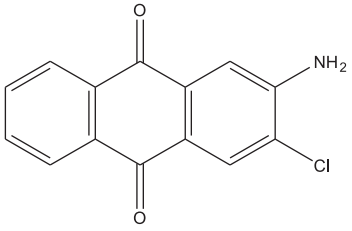
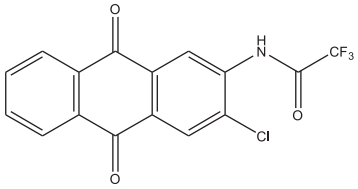
$AQNH_2$		Acid	Time of reaction, h	AQNHC(O)R	
1		2	3	4	
1a		HC(O)OH	6	2a	
		$F_3CC(O)OH$	1	2b	
1b		$F_3CC(O)OH$	1	2c	
1c		HC(O)OH	6	2d	
		$F_3CC(O)OH$	1	2e	

Table continued

1		2	3	4	
1d		HC(O)OH	6	2f	
		$F_3CC(O)OH$	1	2g	
1e		AcOH	3	2h	
		$HSCH_2C(O)OH$	6	2i	
		HC(O)OH	6	2j	
		$F_3CC(O)OH$	1	2k	
1f		$F_3CC(O)OH$	1	2l	

in work [26] on direct acetylation of anilines with ammonium acetate in acetic acid, as well as the experimental data on the absence of reactions when instead of thiocyanate ammonium, thiocyanate potassium or ammonium chloride were used, and direct conversion of 2-amino-9,10-anthraquinone **1e** to amide **2h** in the reaction with the excess of ammonium acetate in acetic acid.

The composition and structure of amides **2a-l** synthesized were confirmed by elemental analysis data, mass spectrometry, and ^{1}H, ^{19}F NMR spectroscopy.

Herein, the new preparative, high yielding method for the synthesis of *N*-acylated amino-9,10-anthraquinones was introduced.

Experimental Part

^{1}H NMR spectra of the compounds synthesized were obtained on a Bruker Avance DRX-500 spectrometer, the internal standard was TMS. ^{19}F NMR spectra were registered on a Varian VXR-300 spectrometer, the internal standard was $CFCl_3$. Chromato-mass spectra were obtained on a Aligent 1100/DAD/HSD/VLG 119,562 device.

The general method of acylation of amino-9,10-anthraquinone 1a-f

To 30 ml of the corresponding carboxylic acid add (0.001 Mol) of amino-9,10-anthraquinone **1a-f**, 0.152 g (0.002 Mol) of ammonium thiocyanate (in the case of compounds **1a,b,f**) or 0.304 g (0.004 Mol) of ammonium thiocyanate (in the case of compounds **1c,d**), and heat when boiling for 1-6 h. Cool the reaction mixture, dilute with the 4-fold excess of water, filter the precipitate, wash with water and dry.

***N*-(9,10-Dioxo-9,10-dihydroanthracen-1-yl)formamide 2a**. Yield – 91%. M.p. – 210-212°C. ^{1}H NMR, δ, ppm.: 8.08-8.17 m (3H, CH_{ar}); 7.87-7.91 m (3H, CH_{ar}); 8.64 м (1H, CH_{ar}); 8.96 br.s (1H, COH); 11.89 br.s (1H, NH). $[M+1]^+$ 252. Found, %: C 71.59; H 3.69; N 5.47. $C_{15}H_9NO_3$. Calculated, %: C 71.71; H 3.61; N 5.58.

***N*-(9,10-Dioxo-9,10-dihydroanthracen-1-yl)-2,2,2-trifluoroacetamide 2b**. Yield – 95%. M.p. – 182-183°C. ^{1}H NMR, δ, ppm.: 7.95-8.24 m (6H, CH_{ar}); 8.74 d (1H, *J*=7.7 Hz, CH_{ar}); 13.17 s (1H, NH). ^{19}F NMR, δ, ppm.: -75.55 (CF_3). $[M+1]^+$ 319. Found, %: C 60.31; H 2.43; N 4.31. $C_{16}H_8F_3NO_3$. Calculated, %: C 60.20; H 2.53; N 4.39.

9,10-Dioxo-1-(2,2,2-trifluoroacetamido)-9,10-dihydroanthracene-2-carboxylic acid 2c. Yield – 92%. M.p. – 287-289°C. ^{1}H NMR, δ, ppm.: 7.77-7.87 m (2H, CH_{ar}); 8.04-8.12 m (4H, CH_{ar}); 11.85 s (1H, NH); 13.03 s (1H, OH). ^{19}F NMR, δ, ppm.: -75.40 (CF_3). $[M+1]^+$ 364. Found, %: C 56.17; H 2.17; N 3.92. $C_{17}H_8F_3NO_5$. Calculated, %: C 56.21; H 2.22; N 3.86.

***N,N'*-(9,10-Dioxo-9,10-dihydroanthracene-1,4-diyl)diformamide 2d**. Yield – 87%. M.p. – 290-292°C. ^{1}H NMR, δ, ppm.: 7.86-8.12 m (5H, CH_{ar}); 8.57 m (1H, CH_{ar}); 8.87 br.s (2H, COH); 12.01 br.s (2H, NH). $[M+1]^+$ 295. Found, %: C 65.40; H 3.36; N 9.57. $C_{16}H_{10}N_2O_4$. Calculated, %: C 65.32; H 3.45; N 9.51.

***N,N'*-(9,10-Dioxo-9,10-dihydroanthracene-1,4-diyl)bis(2,2,2-trifluoroacetamide) 2e**. Yield – 81%. M.p. – 260-261°C. ^{1}H NMR, δ, ppm.: 7.87 m (2H, CH_{ar}); 8.01 m (2H, CH_{ar}); 8.62 m (2H, CH_{ar}); 13.10 br.s (2H, NH). ^{19}F NMR, δ, ppm.: -75.42 (c, $2CF_3$). $[M+1]^+$ 431 Found, %: C 50.35; H 1.81; N 6.58. $C_{18}H_8F_6N_2O_4$. Calculated, %: C 50.25; H 1.87; N 6.51.

***N,N'*-(9,10-Dioxo-9,10-dihydroanthracene-1,5-diyl)diformamide 2f**. Yield – 85%. M.p. > 330°C. ^{1}H NMR, δ, ppm.: 7.86-7.95 m (4H, CH_{ar}); 8.67 m (2H, CH_{ar}); 8.91 br.s (2H, COH); 11.84 br.s (2H, NH). $[M+1]^+$ 295. Found, %: C 65.41; H 3.32; N 9.60. $C_{16}H_{10}N_2O_4$. Calculated, %: C 65.32; H 3.45; N 9.51.

***N,N'*-(9,10-Dioxo-9,10-dihydroanthracene-1,5-diyl)bis(2,2,2-trifluoroacetamide) 2g**. Yield – 79%. M.p. – 242-243°C. ^{1}H NMR, δ, ppm.: 7.67-7.86 m (4H, CH_{ar}); 8.62-8.64 m (2H, CH_{ar}); 12.25 br.s (2H, NH). ^{19}F NMR, δ, ppm.: -75.43 (c, $2CF_3$). $[M+1]^+$ 431. Found, %: C 50.34; H 1.79; N 6.54. $C_{18}H_8F_6N_2O_4$. Calculated, %: C 50.25; H 1.87; N 6.51.

***N*-(9,10-Dioxo-9,10-dihydroanthracen-2-yl)acetamide 2h**. Method A: see the general procedure, yield – 88%. Method B: to 30 ml of acetic acid add 0.223 g (0.001 Mol) of 2-amino-9,10-anthraquinone **1e**, 0.152 g (0.002 Mol) of ammonium acetate and heat when boiling for 3 h. Cool the reaction mixture, dilute with the 4-fold excess of water, filter the precipitate, wash with water and dry. Yield – 87%. M.p. – 258-260°C. ^{1}H NMR, δ, ppm.: 2.12 s (3H, CH_3); 7.88 m (2H, CH_{ar}); 8.04-8.15 m (4H, CH_{ar}); 8.39 s (1H, CH_{ar}); 10.57 s (1H, NH). $[M+1]^+$ 264. Found, %: C 72.54; H 4.01; N 5.32. $C_{16}H_{11}NO_3$. Calculated, %: C 72.45; H 4.18; N 5.28.

***N*-(9,10-Dioxo-9,10-dihydroanthracen-2-yl)-2-mercaptoacetamide 2i**. Yield – 89%. M.p. – 232-233°C. ^{1}H NMR, δ, ppm.: 3.04 s (1H, SH); 3.81 m (2H, CH_2); 7.81-7.87 m (3H, CH_{ar}); 8.12 m (3H, CH_{ar}); 8.35 s (1H, CH_{ar}); 10.70 s (1H, NH).: $[M+1]^+$ 297. Found, %: C 64.74; H 3.59; N 4.82; $C_{16}H_{11}NO_3S$. Calculated, %: C 64.63; H 3.73; N 4.71.

***N*-(9,10-Dioxo-9,10-dihydroanthracen-2-yl)formamide 2j**. Yield – 93%. M.p. – 282-283°C. ^{1}H NMR, δ, ppm.: 7.84-7.87 m (2H, CH_{ar}); 7.96-7.99 m (1H, CH_{ar}); 8.08-8.11 m (3H, CH_{ar}); 8.36 s (1H, COH); 8.41 s (1H, CH_{ar});10.79 s (1H, NH). $[M+1]^+$ 252. Found, %: C 71.56; H 3.67; N 5.50. $C_{15}H_9NO_3$. Calculated, %: C 71.71; H 3.61; N 5.58.

***N*-(9,10-Dioxo-9,10-dihydroanthracen-2-yl)-2,2,2-trifluoroacetamide 2k**. Yield – 96%. M.p. – 216-217°C. ^{1}H NMR, δ, ppm.: 8.13-8.36 m (4H, CH_{ar}); 8.75-7.81 m (3H, CH_{ar}); 13.01 s (1H, NH). ^{19}F NMR, δ, ppm.: -75.61 c (CF_3). $[M+1]^+$ 319. Found, %: C 60.32; H 2.48; N 4.29. $C_{16}H_8F_3NO_3$. Calculated, %: C 60.20; H 2.53; N 4.39.

***N*-(3-Chloro-9,10-dioxo-9,10-dihydroanthracen-2-yl)-2,2,2-trifluoroacetamide 2l**. Yield – 90%. M.p. > 330°C. ^{1}H NMR, δ, ppm.: 7.90-8.25 m (6H, CH_{ar}); 11.69 br.s (1H, NH). ^{19}F NMR, δ, ppm.: -75.16 c, (CF_3). $[M+1]^+$ 354. Found, %: C 54.27; H 2.10; Cl 10.12; N 3.91. $C_{16}H_7ClF_3NO_3$. Calculated, %: C 54.34; H 2.00; Cl 10.02; N 3.96.

Conclusions

The effective method for the synthesis of *N*-formyl(acetyl-, mercaptoacetyl- and trifluoroacetyl)amino-9,10-anthraquinones based on the interaction of amino-9,10-anthraquinones with the corresponding carboxylic acids in the presence of the excess ammonium thiocyanate has been developed.

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THE SYNTHESIS AND THE ANTITUBERCULAR ACTIVITY OF 1-BENZYL-4-HYDROXY-2-OXO-1,2,5,6,7,8-HEXAHYDROQUINOLINE-3-CARBOXAMIDES

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Key words: amides; 4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxylic acids; synthesis; thermolysis; antitubercular activity

Unfortunately, tuberculosis still remains a cause of high mortality in humans and in modern conditions it has become a global health problem. Continuing the search for new antimycobacterial agents among the amidated derivatives of 4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxylic acids the corresponding group of 1-benzyl-4-hydroxy-2-oxo-1,2,5,6,7,8-hexahydroquinoline-3-carboxamides has been synthesized by the reaction of ethyl 1-benzyl-4-hydroxy-2-oxo-1,2,5,6,7,8-hexahydroquinoline-3-carboxylate and anilines or hetarylamines in DMF at 130°C. The chemical structure of the compounds obtained has been confirmed by the data of elemental analysis, NMR ^{1}H spectroscopy and mass spectrometry. It has been noted that the 1H NMR spectra can reliably confirm the presence of the basic functional groups by their corresponding chemical shift, the integrated intensity and multiplicity of signals. It has been shown that under the influence of the electron impact the molecular ions of all the compounds studied undergo the primary fragmentation in two directions: with breaking the amide bond or the quinolone nucleus – the carbamide moiety bond. According to the data of microbiological tests among 1-benzyl-4-hydroxy-2-oxo-1,2,5,6,7,8-hexahydroquinoline-3-carboxamides synthesized the substances that are capable of inhibiting actively the growth of Mycobacterium tuberculosis H37Rv in low concentration have been identified, and therefore, they are of interest for further research.

СИНТЕЗ ТА ПРОТИТУБЕРКУЛЬОЗНА АКТИВНІСТЬ 1-БЕНЗИЛ-4-ГІДРОКСИ-2-ОКСО-1,2,5,6,7,8-ГЕКСАГІДРОХІНОЛІН-3-КАРБОКСАМІДІВ

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Ключові слова: *аміди; 4-гідрокси-2-оксо-1,2-дигідрохінолін-3-карбонові кислоти; синтез; термоліз; протитуберкульозна активність*

На жаль, туберкульоз до сьогодні залишається причиною високої смертності людей і в сучасних умовах перетворився на глобальну проблему системи охорони здоров'я. Продовжуючи пошук нових антимікобактеріальних засобів серед амідованих похідних 4-гідрокси-2-оксо-1,2-дигідрохінолін-3-карбонових кислот, реакцією етил 1-бензил-4-гідрокси-2-оксо-1,2,5,6,7,8-гексагідрохінолін-3-карбоксилату з анілінами чи гетариламінами в ДМФА при 130°С було синтезовано групу відповідних 1-бензил-4-гідрокси-2-оксо-1,2,5,6,7,8-гексагідрохінолін-3-карбоксамідів. Хімічна будова одержаних сполук була підтверджена даними елементного аналізу, спектроскопії ЯМР ^{1}H та мас-спектрометрії. Відмічено, що ЯМР ^{1}H спектри дозволяють надійно підтвердити присутність усіх основних функціональних груп за відповідними їм за хімічним зсувом, інтегральною інтенсивністю та мультиплетністю сигналами. Показано, що під впливом електронного удару молекулярні іони всіх досліджуваних сполук піддаються первинній фрагментації за двома напрямками: з розривом амідного зв'язку чи зв'язку хінолонове ядро – карбамідний фрагмент. За даними мікробіологічних випробовувань серед синтезованих 1-бензил-4-гідрокси-2-оксо-1,2,5,6,7,8-гексагідрохінолін-3-карбоксамідів виявлені речовини, здатні в низькій концентрації активно затримувати ріст Mycobacterium tuberculosis H37Rv і тому представляють інтерес для подальших досліджень.

СИНТЕЗ И ПРОТИВОТУБЕРКУЛЕЗНАЯ АКТИВНОСТЬ 1-БЕНЗИЛ-4-ГИДРОКСИ-2-ОКСО-1,2,5,6,7,8-ГЕКСАГИДРОХИНОЛИН-3-КАРБОКСАМИДОВ

А.А.Давиденко

Ключевые слова: *амиды; 4-гидрокси-2-оксо-1,2-дигидрохинолин-3-карбоновые кислоты; синтез; термолиз; противотуберкулезная активность*

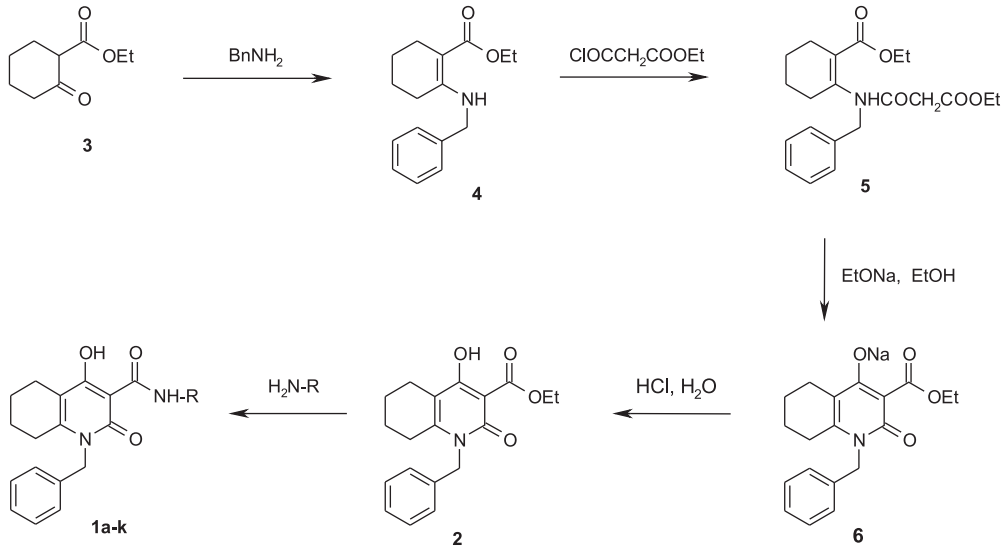
К сожалению, туберкулез до настоящего времени остается причиной высокой смертности людей и в современных условиях превратился в глобальную проблему здравоохранения. Продолжая поиск новых антимикобактериальных средств среди амидированных производных 4-гидрокси-2-оксо-1,2-дигидрохинолин-3-карбоновых кислот, реакцией этил 1-бензил-4-гидрокси-2-оксо-1,2,5,6,7,8-гексагидрохинолин-3-карбоксилата с анилинами или гетарил-аминами в ДМФА при 130°С была синтезирована группа соответствующих 1-бензил-4-гидрокси-2-оксо-1,2,5,6,7,8-гексагидрохинолин-3-карбоксамидов. Химическое строение полученных соединений было подтверждено данными элементного анализа, спектроскопии ЯМР ^{1}H и масс-спектрометрии. Отмечено, что ЯМР ^{1}H спектры позволяют надежно подтвердить присутствие всех основных функциональных групп по соответствующим им по химическому сдвигу, интегральной интенсивности и мультиплетности сигналам. Показано, что под воздействием электронного удара молекулярные ионы всех изучаемых соединений претерпевают первичную фрагментацию по двум направлениям: с разрывом амидной связи или связи хинолоновое ядро – карбамидный фрагмент. По данным микробиологических испытаний среди синтезированных 1-бензил-4-гидрокси-2-оксо-1,2,5,6,7,8-гексагидрохинолин-3-карбоксамидов обнаружены вещества, способные в низкой концентрации активно ингибировать рост Mycobacterium tuberculosis H37Rv и поэтому представляющие интерес для дальнейших исследований.

Despite the considerable progress and success of modern medicine in the fight against many infectious diseases tuberculosis remains one of the main causes of the high mortality. Unfortunately, in recent years this dangerous disease has not only returned, but is also extremely widespread throughout the world, becoming a global public health problem. One of the main causes for this situation was the unique ability of the causative agent – *Mycobacterium tuberculosis* – to very rapid mutations and, as a result, there is immediate distribution of strains that are resistant to the existing medicines [1-2]. Treatment of such patients is very difficult, time consuming, with substantial financial costs and, moreover, is not always successful. Therefore, in the present circumstances the fight against tuberculosis is carried out in several directions simultaneously. Thus, reliable methods of diagnosis, which allow determining the true pathogen rapidly and accurately, and its sensitivity to drugs are being developed, and it gives the possibility to start treatment promptly and optimally [3-6]. Decoding of the genome of *Mycobacterium tuberculosis* and searching for the genes responsible for production of drug resistance are very interesting [7]. The search of completely new biologically active substances of various chemical classes that can effectively inhibit the growth of *Mycobacterium tuberculosis* at all stages of development do not lose its relevance [8-11].

In this regard 1-R-4-hydroxy-2-oxo-1,2,5,6,7,8-hexahydroquinoline-3-carboxamides are of undoubted interest. Among them the highly active anti-tuberculosis agents were previously found. At the same time it is repeatedly indicated that the biological activity of these compounds is largely determined by the nature of the substituent at the cyclic nitrogen atom of the quinoline nucleus [12-14]. Anilides and hetarylamides of 1-furfuryl-4-hydroxy-2-oxo-1,2,5,6,7,8-hexahydroquinoline-3-carboxylic acids were indicated as promising objects [15]. Continuing research in this area the replacement of the furan cycle in the structure of the 1-N-substituent with the phenyl nucleus close to it by its structure and properties is interesting. The methodology of bioisosteric replacements widely and successfully used in modern medicinal chemistry has become the theoretical underpinning for this modification, i.e. replacing one of the fragments of the molecule with another one having similar physical and chemical characteristics and inducing a similar biological effect [16-18].

The synthesis of the target objects of research – 1-benzyl-4-hydroxy-2-oxo-1,2,5,6,7,8-hexahydroquinoline-3-carboxamides (**1a-k**) was carried out by the reaction of ethyl ester (**2**) and the corresponding primary amines in thermolysis conditions (Scheme 1). Only those anilines and hetarylamines that had already proven themselves as an excellent base for highly active anti-tuberculosis agents were involved in the synthesis. In turn, ester (**2**) was prepared from commercially available 2-oxocyclohexanecarboxylic acid ethyl ester (**3**) easily forming enamine with benzylamine (**4**). The subsequent acylation by ethyl malonyl chloride gives diester (**5**), which intramolecular condensation leads first to sodium salt (**6**) and further to the initial ester (**2**).

The 1-benzyl-4-hydroxy-2-oxo-1,2,5,6,7,8-hexahydroquinoline-3-carboxamides (1a-k) obtained are colourless crystalline substances with the narrow range of melting points, when heating they are readily soluble in DMF and DMSO, sparingly soluble in alcohol, and practically insoluble in water (Table 1). Their structure was confirmed by elemental analysis, ^{1}H NMR spectra and mass spectra.



1 a R = 3-F-C_6H_4; **b** R = 4-F-C_6H_4; **c** R = 3-Cl-C_6H_4; **d** R = 2,4-$(Cl)_2$-C_6H_3; **e** R = 3-Br-C_6H_4; **f** R = 3-Py; **g** R = 1,3-thiazole-2-yl; **h** R = 4-(adamantane-1-yl)-1,3-thiazole-2-yl; **i** R = bebnzothiazole-2-yl; **j** R = 6-bromobebnzothiazole-2-yl; **k** R = 5-methyl-1,3,4-thiadiazole-2-yl

Scheme 1

Table 1

Characteristics of amides (1a-k)

Compound	Empirical formula	Found, % / Calculated, %			Mp, °C	Yield, %	Antitubercular activity*
		C	H	N			
1a	$C_{23}H_{21}FN_2O_3$	70.33 / 70.40	5.48 / 5.39	7.20 / 7.14	131-133	88	13
1b	$C_{23}H_{21}FN_2O_3$	70.31 / 70.40	5.45 / 5.39	7.06 / 7.14	147-149	94	63
1c	$C_{23}H_{21}ClN_2O_3$	67.68 / 67.56	5.11 / 5.18	6.93 / 6.85	126-128	90	28
1d	$C_{23}H_{20}Cl_2N_2O_3$	62.22 / 62.31	4.64 / 4.55	6.42 / 6.32	135-137	85	7
1e	$C_{23}H_{21}BrN_2O_3$	61.02 / 60.94	4.58 / 4.67	6.25 / 6.18	150-152	92	8
1f	$C_{22}H_{21}N_3O_3$	70.47 / 70.38	5.56 / 5.64	11.24 / 11.19	144-146	91	90
1g	$C_{20}H_{19}N_3O_3S$	63.07 / 62.98	4.96 / 5.02	10.95 / 11.02	141-143	87	94
1h	$C_{30}H_{33}N_3O_3S$	69.79 / 69.88	6.53 / 6.45	8.08 / 8.15	165-167	95	10
1i	$C_{24}H_{21}N_3O_3S$	66.88 / 66.80	4.85 / 4.91	9.81 / 9.74	159-161	90	66
1j	$C_{24}H_{20}BrN_3O_3S$	56.56 / 56.48	4.02 / 3.95	8.14 / 8.23	168-170	93	39
1k	$C_{20}H_{20}N_4O_3S$	60.64 / 60.59	5.16 / 5.08	14.05 / 14.13	143-145	86	97

* – The growth inhibition (%) of *Mycobacterium tuberculosis* H37Rv ATCC 27294 in the concentration of 6.25 μg/ml.

All main functional groups of amides (**1a-k**) containing protons in ^{1}H NMR spectra are identified without complications (Table 2). For example, the protons of 4-OH groups are evident as singlets with integrated intensity of 1H in the typically weak field range (15.63-14.11 ppm) as should be expected for enolic hydroxyls. Singlets of protons of amide groups are regularly slightly shifted upfield: 13.94-12.54 ppm. Further there is the "aromatic" area, in which protons of the phenyl nucleus of the 1-N-benzyl substituent resonate, and aromatic protons of anilide and hetarylamide fragments. The methylene bridge separating the cyclic nitrogen atom and the phenyl nucleus is evident as a singlet with the intensity of 2H in a relatively strong field: 5.42-5.38 ppm. Methylene units of the hexahydroquinoline bicycle have the form of narrow multiplets in the strongest field of the spectrum, wherein if the chemical shifts 8-CH_2 and 5-CH_2-groups are significantly different, the resonant frequency of 6-CH_2 and 7-CH_2-groups are so close that they are almost impossible to be distinguished (Table 2).

An important and useful information on the structure of 1-benzyl-4-hydroxy-2-oxo-1,2,5,6,7,8-hexahydroquinoline-3-carboxamides (**1a-k**) is provided by their mass spectra (Table 2). So, all the compounds synthesized under the influence of electron impact form peaks of molecular ions of low intensity and multiplicity corresponding to the isotopic composition [19]. The primary fragmentation of molecular ion occurs in two directions with approximately equal probability. The first of them is C(3)–CONHHet(Ar) bond breaking or the pathway A (Scheme 2), the result is formation of fragment ions of isocyanate **7** and benzylquinoline **8**. The second direction is destruction of the molecular radical cation by the carbamide bond (pathway B), it is the cause of appearance of highly intense peaks of the released amines (2-aminobenzothiazole 9 in case of amide **1i**) in the spectra and, although it is less intense, but it is common for all compounds of the fragment ion of ketene **10** with m/z 281. It is interesting that the loss of the 1-N-benzyl substituent occurs only during the secondary fragmentation, and it is not observed in any of the examples studied in the primary decomposition of the molecular ion.

The anti-tuberculosis activity of 1-benzyl-4-hydroxy-2-oxo-1,2,5,6,7,8-hexahydroquinoline-3-carboxamides (**1a-k**) was studied by the radiometric method [20, 21]. A comparative analysis of the experimental data (Table 1) obtained with the results of

Table 2

^{1}H NMR and mass spectra of amides (1a-k)

Compound	^{1}H NMR spectra, Chemical shifts, δ, ppm (*J*, Hz)							Mass spectra, $[M]^+$ (I_{rel}, %)
	4-OH (1H, s)	NH (1H, s)	1N-CH_2 (2H, s)	Hexahydroquinoline nucleus			Proton signals of other functional groups	
				8-CH_2 (2H, m)	5-CH_2 (2H, m)	6,7-$(CH_2)_2$ (4H, m)		
1a	15.58	12.61	5.40	2.66	2.41	1.65	7.63-6.96 (9H, m, Ph + H arom.)	392 (14)
1b	15.63	12.65	5.38	2.67	2.42	1.64	7.69-7.11 (9H, m, Ph + H arom.)	392 (17)
1c	15.57	12.54	5.41	2.66	2.40	1.67	7.82-7.10 (9H, m, Ph + H arom.)	408/410 (12/3)
1d	15.22	12.98	5.39	2.71	2.41	1.69	8.33-7.32 (8H, m, Ph + H arom.)	442/444/446 (9/6/1)
1e	15.34	12.79	5.40	2.68	2.40	1.68	7.52-7.12 (9H, m, Ph + H arom.)	452/454 (13/12)
1f	15.41	12.77	5.39	2.69	2.40	1.70	8.80 (1H, s, 2-H Py); 8.35 (1H, d, *J* = 4.4, 6-H Py); 8.10 (1H, d, *J* = 8.0, 4-H Py); 7.43-7.12 (6H, m, 5-H Py + Ph)	375 (18)
1g	14.52	13.63	5.38	2.70	2.41	1.69	7.41-7.05 (7H, m, Ph + 4,5-H Th)	381 (15)
1h	14.46	13.68	5.40	2.70	2.40	1.64	7.33-7,12 (5H, m, Ph); 6.88 (1H, s, 5-H Th); 2.00 (3H, s, γ-H Ad); 1.84 (6H, s, δ-H Ad); 1.76 (8H, s, β-H Ad)	515 (4)
1i	14.17	13.92	5.42	2.66	2.39	1.65	7.91-7.10 (9H, m, Ph + H arom.)	431 (10)
1j	14.00	13.89	5.41	2.67	2.40	1.66	8.20 (1H, s, 7′-H); 7.70-7.11 (7H, m, Ph + H arom.)	509/511 (11/12)
1k	14.11	13.94	5.40	2.66	2.41	1.67	7.40-7.12 (5H, m, Ph); 2.72 (3H, s, Me)	396 (19)

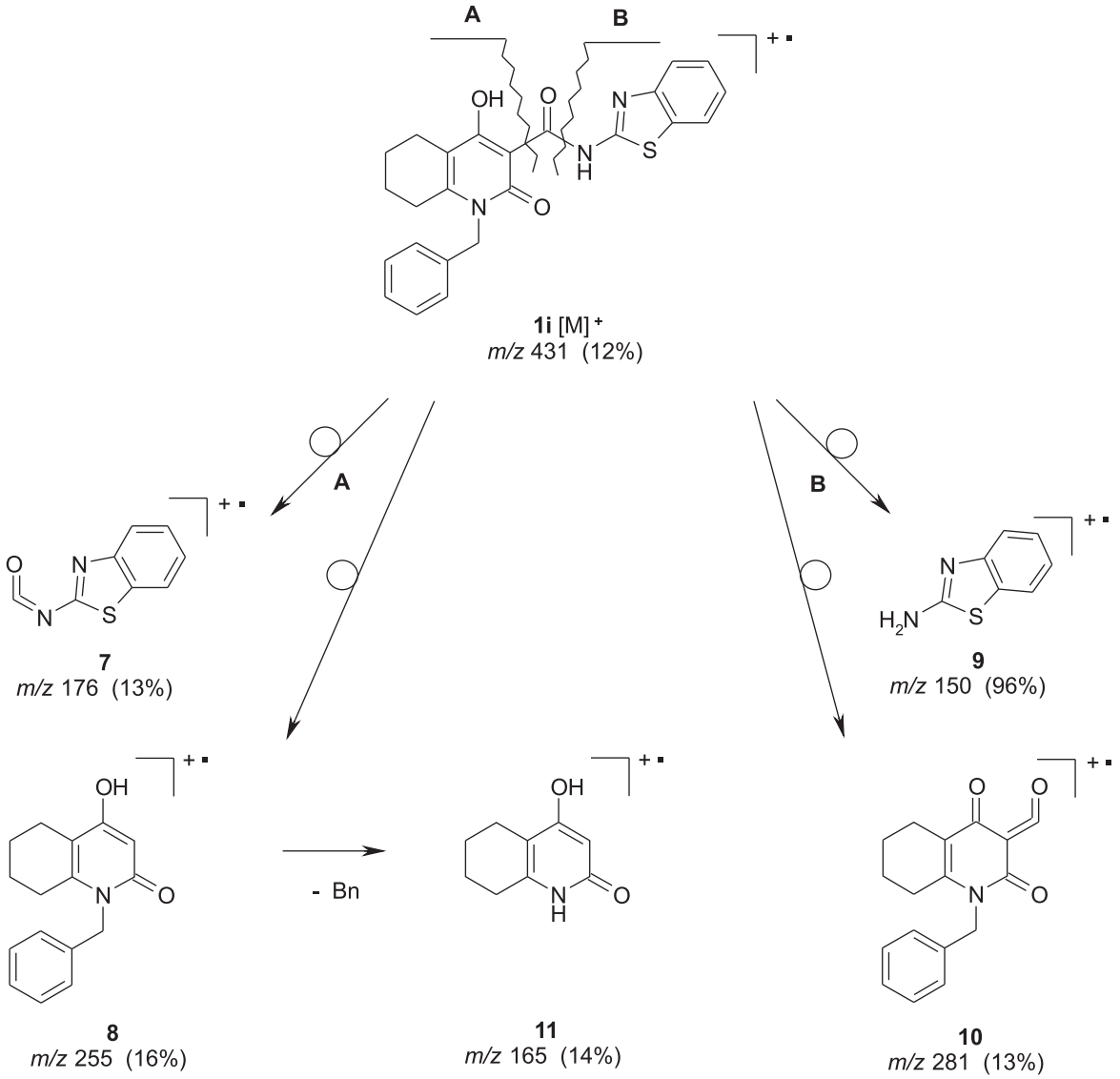
1-N-furfuryl-substituted derivatives with the similar structure shown in similar conditions was performed [15]. It has been found that in the vast majority of cases the replacement of the furan ring with the benzene ring is accompanied by a significant increase in antimycobacterial properties and, therefore, is a good variant of a chemical modification.

The most active compounds of this series – pyridine-3-ylamide 1f, 1,3-thiazole-2-ylamide **1g** and 5-methyl-1,3,4-thiadiazole-2-ylamide **3k** – in the concentration of 6.25 μg/mL can inhibit the growth of *Mycobacterium tuberculosis* H37Rv ATCC 27294 by more than 90%, and thus, they are transferred to the next stage of the microbiological testing as promising antimycobacterial agents.

Experimental Part

The ^{1}H NMR spectra were recorded on a Varian Mercury-400 spectrometer (400 MHz) in DMSO-d_6 solution, the internal standard was TMS. Mass spectra were recorded of a Varian 1200L instrument in full scan mode in the range of 35-700 *m/z*, with EI ionization (70 eV) and direct sample introduction. Elemental analysis was carried out on a EuroVector EA-3000 microanalyzer. Melting points were determined in capillaries on a SMP10 Stuart digital melting point analyzer. The commercial 2-oxocyclohexanecarboxylic acid ethyl ester, benzylamine and ethoxymalonyl chloride of Fluka company were used in the synthesis.

Ethyl 1-benzyl-4-hydroxy-2-oxo-1,2,5,6,7,8-hexahydroquinoline-3-carboxylate (2). Stir vigorously the mixture of 17.02 g (0.1 Mol) 2-oxocyclohexanecarboxylic acid ethyl ester (**3**) and 12.00 mL (0.11 Mol) of benzylamine at room temperature for 1 h. Heat the reaction mixture to 50°C, and continue to stir for 5 h, then remove the excess of benzylamine in vacuum. Dissolve the residue 200 mL of CH_2Cl_2. Distill off about 50 ml of the solvent, while removing azeotropically the water formed during the reaction of cyclohexanone **3** with benzylamine. To the solution of the enamine **4** obtained in CH_2C_{l2} add at first 14 mL (0.1 Mol) of triethylamine and then while stirring and cooling 15.05 g (0.1 Mol) of ethoxymalonyl chloride. In 5-6 h add 300 mL of cold water to the reaction mixture and mix thoroughly. Separate the organic layer, dry over anhydrous $CaCl_2$. At the end distill the solvent under reduced pressure. To the residue (amido ester **5**) add the solution of sodium ethoxide [4.6 g (0.2 Mol) of sodium metal and 150 mL of absolute ethanol], heat to boiling and allow to stand for 3 h at room temperature. Add 500 mL of cold water, acidify with HCl to pH4. Filter the precipitate of ester **2** formed, wash with cold water and dry. Yield – 26.5 g (81%). M.p. – 87-89°C (EtOH). ^{1}H NMR spectrum, δ, ppm, (*J*, Hz): 13.32 (1H, s, OH); 7.33 (2H, t, *J* = 7.8, 3,5-H Ph); 7.24 (1H, t, *J* = 7.6, 4-H Ph); 7.10 (2H, d, *J* = 8.1, 2,6-H Ph); 5.26 (2H, s, N-CH_2); 4.32 (2H, q, *J* = 7.0, O$\underline{C}H_2CH_3$); 2.62 (2H, m, 8-CH_2); 2.43 (2H, m, 5-CH_2); 1.62 (4H, m, 6,7-CH_2); 1.30 (3H, t, *J* = 7.0, $OCH_2C\underline{H}_3$). Found, %:



Scheme 2

C 69.80; H 6.53; N 4.21. $C_{19}H_{21}NO_4$. Calculated, %: C 69.71; H 6.47; N 4.28.

1-Benzyl-4-hydroxy-2-oxo-1,2,5,6,7,8-hexahydroquinoline-3-carboxamides (1a-k). The general procedure. Stir the mixture of 3.27 g (0.01 Mol) of ethyl ester **2**, the corresponding aniline or hetarylamine (0.01 Mol) and 2 ml of DMF and allow to stand on a metal bath at 130°C for 5 min. Cool the reaction mixture, add 10 ml of ethanol and triturate thoroughly. Filter the amide **1a-k** precipitated, wash with alcohol, dry, and recrystallize from the mixture DMF and EtOH.

Conclusions

1. The preparative method for obtaining the substances has been proposed, and the synthesis of a new series of 1-benzyl-4-hydroxy-2-oxo-1,2,5,6,7,8-hexahydroquinoline-3-carboxamides has been carried out. Their structure has been confirmed by elemental analysis, ^{1}H NMR spectra and mass spectra.

2. According to the data of the microbiological testing some substances exhibiting a high anti-tuberculosis activity in low concentrations have been identified and recommended for *in vivo* studies in the range of the compounds studied.

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THE SYNTHESIS, COMPUTER PREDICTION OF THE BIOLOGICAL ACTIVITY AND THE ACUTE TOXICITY OF 1-Ar-4-*R*-[1,2,4] TRIAZOLO[4,3-*a*] QUINAZOLIN-5(4*H*)-ONES

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Key words: computer prediction; biological activity; acute toxicity; synthesis; 2-hydrazinoquinazolin-4(3H)-ones; [1,2,4]triazolo[4,3-a]quinazolin-5(4H)-ones

Using the PASS programme computer prediction of the biological activity of 1-Ar-4-R-[1,2,4]triazolo[4,3-a] quinazolin-5(4H)-ones has been performed; it has allowed to identify the types of the biological activity of the compounds studied and sort out the most promising compounds 5{1-20} with the potential anti-asthmatic and anti-allergic activity. Prediction of the acute toxicity of 1-Ar-4-R-[1,2,4]triazolo[4,3-a]quinazolin-5(4H)-ones 5{1-20} has been carried out by the GUSAR software, which allows to refer them to slightly toxic (toxicity class 4) or practically nontoxic (toxicity class 5) substances. The synthesis of the most promising compounds 5{1-20} studied in silico for the biological activity and the acute toxicity has been conducted by interaction of the corresponding 2-hydrazinoquinazolin-4(3H)-ones 1{1-5} with imidazolides 3{1, 2} of aromatic acids 2{1, 2}, or with aromatic aldehydes 4{1, 2} followed by oxidation in the presence of $FeCl_3$. The structure of the compounds 5{1-20} synthesized has been proven by the data of the elemental analysis and ^1H NMR spectroscopy. The compounds obtained are promising objects for further investigations as slightly toxic or nontoxic substances with the potential anti-asthmatic and anti-allergic activity.

СИНТЕЗ, КОМП'ЮТЕРНЕ ПРОГНОЗУВАННЯ БІОЛОГІЧНОЇ АКТИВНОСТІ ТА ГОСТРОЇ ТОКСИЧНОСТІ 1-Ar-4-R-[1,2,4]ТРИАЗОЛО[4,3-a]ХІНАЗОЛІН-5(4H)-ОНІВ
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Ключові слова: *комп'ютерне прогнозування; біологічна активність; гостра токсичність; синтез; 2-гідразинохіназолін-4(3H)-они; [1,2,4]триазоло-[4,3-a]хіназолін-5(4H)-они*
Проведено комп'ютерне прогнозування біологічної активності віртуальної бібліотеки 1-Ar-4-R-[1,2,4] триазоло[4,3-a]хіназолін-5(4H)-онів за допомогою комп'ютерної програми PASS, що дозволило визначити напрямок біологічної активності досліджуваних сполук та виділити серед них найбільш перспективні 5{1-20} з потенційною протиастматичною та антиалергічною активністю. Комп'ютерне прогнозування гострої токсичності 1-Ar-4-R-[1,2,4]триазоло[4,3-a]хіназолін-5(4H)-онів 5{1-20} здійснено за програмним забезпеченням GUSAR, що дозволило віднести їх до малотоксичних (4 клас токсичності) або практично нетоксичних речовин (5 клас токсичності). Синтез найбільш перспективних сполук 5{1-20}, досліджених методом in silico на біологічну активність та гостру токсичність, був проведений при взаємодії відповідних 2-гідразинохіназолін-4(3H)-онів 1{1-5} з імідазолідами 3{1, 2} ароматичних кислот 2{1, 2} або з ароматичними альдегідами 4{1, 2} з наступним окисненням у присутності $FeCl_3$. Будову синтезованих сполук 5{1-20} доведено за допомогою елементного аналізу та даних ^1H ЯМР спектроскопії. Отримані сполуки є перспективними об'єктами для подальших досліджень як малотоксичні або нетоксичні речовини з потенційною протиастматичною та антиалергічною активністю.

СИНТЕЗ, КОМПЬЮТЕРНОЕ ПРОГНОЗИРОВАНИЕ БИОЛОГИЧЕСКОЙ АКТИВНОСТИ И ОСТРОЙ ТОКСИЧНОСТИ 1-Ar-4-R-[1,2,4]ТРИАЗОЛО[4,3-a]ХИНАЗОЛИН-5(4H)-ОНОВ
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Ключевые слова: *компьютерное прогнозирование; биологическая активность; острая токсичность, синтез; 2-гидразинохиназолин-4(3H)-оны; [1,2,4]триазоло[4,3-a]хиназолин-5(4H)-оны*
Проведено компьютерное прогнозирование биологической активности виртуальной библиотеки 1-Ar-4-R-[1,2,4]триазоло[4,3-a]хиназолин-5(4H)-онов с помощью компьютерной программы PASS, что позволило определить направление биологической активности исследуемых соединений и выделить из них наиболее перспективные 5{1-20} с потенциальной противоастматической и антиаллергической активностью. Компьютерное прогнозирование острой токсичности 1-Ar-4-R-[1,2,4]триазоло[4,3-a] хиназолин-5(4H)-онов 5{1-20} проведено за счет использования программного обеспечения GUSAR, что позволило отнести их к малотоксичным (4 класс токсичности) или практически нетоксичным веществам (5 класс токсичности). Синтез наиболее перспективних соединений 5{1-20}, исследованных методом in silico на биологическую активность и острую токсичность, был проведен при взаимодействии соответствующих 2-гидразинохиназолин-4(3H)-онов 1{1-5} с имидазолидами 3{1, 2} ароматических кислот 2{1, 2} или с ароматическими альдегидами 4{1, 2} с последующим окислением в присутствии $FeCl_3$. Строение синтезированных соединений 5{1-20} доказано при помощи элементного анализа и данных ^1H ЯМР-спектроскопии. Полученные соединения являются перспективными объектами для дальнейших исследований как малотоксичные или нетоксичные вещества с потенциальной противоастматической и антиаллергической активностью.

In recent years domestic and foreign researchers pay much attention to the targeted synthesis of low toxic compounds with the expressed biological properties, and it is an important stage in development of innovative drug substances. Derivatives of [1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-one, which are representatives of the important class of condensed heterocycles possessing a wide range of the biological activity, attract particular interest in this direction. Among their potential pharmacologically significant properties the H1-antihistaminic [1-11], anticonvulsant [12], antibacterial [13-15], antitubercular [13, 15], antifungal [13, 14], anticancer [15], anti-asthmatic [10, 16], antiHIV [13], anti-allergic [16], anti-inflammatory [16, 17] bioactivities should be mentioned. It determines the prospects for developing synthetic approaches to fundamentally new compounds of the specified class.

The possibility to synthesize a large amount of [1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-one derivatives leads to understanding of the necessity for the rational presynthetic selection of the most promising compounds of this class. One of the effective ways to solve this problem is computer prediction of various properties of [1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-one derivatives, such as the biological activity [18] and the acute toxicity [19]; it allows to eliminate unpromising substances at the early stages of the research.

Taking into account the actuality of searching biological active substances among [1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-one derivatives and modern advances in software for virtual screening the goal was to conduct modelling of the virtual library of 1-Ar-4-R-[1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-ones, to determine the most probable biological activity spectrum and the acute toxicity of the compounds studied using the PASS and GUSAR software, sort out the most promising substances and develop preparative methods for their synthesis.

Results and Discussion

For design of the virual library of 1-Ar-4-R-[1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-ones two randomization points in position 1 and 4 of [1,2,4]triazolo-[4,3-*a*]quinazolin-5(4*H*)-one were chosen.

Analysis of the computer prediction results for the virual library of 1-Ar-4-R-[1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-ones by the PASS software showed the possibility of searching substances possessing the anti-asthmatic and anti-allergic activity among these compounds and allowed to generate the library of the most promising compounds **5***{1-20}* for further research (Table 1) [18].

The data of computer prediction of the biological activity obtained are fully consistent with the fact that [1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-one derivatives are similar in their chemical structure to the chemical structures described as promising non-sedative H1-antihistaminic drugs [1-11].

The research results *in silico* by the GUSAR software gave the possbility to predict the acute toxicity values for different routes of administration of 1-Ar-4-*R*-[1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-ones **5***{1-20}* (Table 2). The values of LD_{50} in the intraperitoneal administration were between 391 to 960 mg/kg, when introducing intravenously they were between 91 to 251 mg/kg. The values of LD_{50} in the oral administration were between 556 to 1891 mg/kg, and when introducing subcutaneously – 405 to 2934 mg/kg [19]. The data obtained indicate that compounds **5***{1-20}* are slightly toxic (toxicity class 4) or practically non-toxic (toxicity class 5) [19, 20].

The synthesis of 1-Ar-4-*R*-[1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-ones without hydroxyls **5***{1, 4, 5, 8, 9, 12, 13, 16, 17, 20}* was carried out by interaction of the corresponding 2-hydrazinoquinazolin-4(3*H*)-ones **1***{1-5}* previously synthesized according to the improved method [21] with imidazolides **3***{1, 2}* of aromatic acids **2***{1, 2}* preliminary obtained via carbonyldiimidazole (CDI). This way allows obtaining the final products in good yields, but is not suitable for hydroxyl-containing compounds due to adverse reactions. Hydroxyl-containing compounds **5***{2, 3, 6, 7, 10, 11, 14, 15, 18, 19}* were synthesized by the reaction of 2-hydrazinoquinazolin-4(3*H*)-ones **1***{1-5}* with aromatic aldehydes **4***{1, 2}* followed by oxidation in the presense of $FeCl_3$ (Scheme).

The structures of the compounds **5***{1-20}* obtained were confirmed by the ^{1}H NMR spectroscopy data (Table 3). Formation of the [1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-ones condenced system leads to shift of H-6 protons signals to 8.22-8.26 ppm, and it is in good correlation with the known data [22].

Experimental Part

The virtual screening for the biological activity of the virtual library of the substances studied was performed by the PASS Online web-resource. It enables to predict more than 4000 types of the biological activity with the average accuracy of more than 95% based on the analysis of the structure – activity relationships in a training set (drug substances, drug candidates being at various stages of clinical or preclinical trials, pharmacological substances and biochemical reagents, substances with the known specific toxicity data), which contains information about the structure and the biological activity of more than 300000 organic compounds [23-25].

Computer prediction of the biological activity spectrum of the virtual library of [1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-one derivatives was performed with probability of demonstration of the specific type of the therapeutic action exceeding 50% (Pa>0.500). It al-

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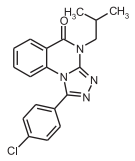
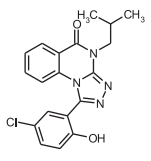
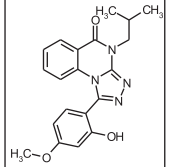
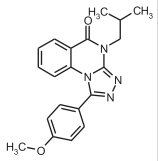
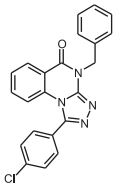
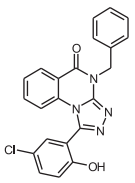
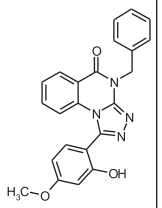
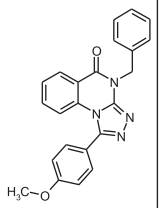
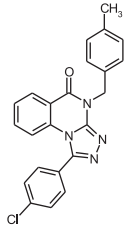
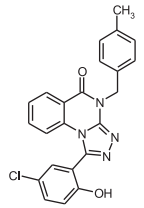
Table 1

Prediction of the biological activity spectrum of 1-Ar-4-R-[1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-ones **5***{1-10}* [18]

Biological activity	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi
	5*{1}*		**5***{2}*		**5***{3}*		**5***{4}*		**5***{5}*		**5***{6}*		**5***{7}*		**5***{8}*		**5***{9}*		**5***{10}*	
																				
Anti-asthmatic	0.639	0.012	0.573	0.018	0.599	0.016	0.651	0.011	0.698	0.009	0.617	0.014	0.643	0.012	0.696	0.009	0.640	0.012	0.573	0.018
Anti-allergic	0.629	0.013	0.590	0.016	0.615	0.014	0.644	0.012	0.662	0.011	0.609	0.015	0.636	0.013	0.662	0.011	0.613	0.014	0.577	0.018
Tumour necrosis factor alpha release inhibitor	–	–	–	–	–	–	0.517	0.008	0.579	0.005	–	–	0.532	0.007	0.626	0.005	0.528	0.007	–	–
CYP2A8 substrate	–	–	–	–	–	–	–	–	0.523	0.029	–	–	–	–	–	–	–	–	–	–
Tetrahydroxynaphthalene reductase inhibitor	–	–	–	–	–	–	–	–	–	–	0.589	0.009	–	–	–	–	–	–	0.522	0.013
5-O-(4-coumaroyl)-D-quinate 3′-monooxygenase inhibitor	–	–	–	–	–	–	–	–	0.527	0.073	–	–	–	–	–	–	–	–	–	–
Glycosylphosphatidylinositol phospholipase D inhibitor	–	–	–	–	–	–	–	–	0.512	0.095	–	–	–	–	–	–	–	–	–	–
Hepatic function stimulant	–	–	–	–	–	–	–	–	0.500	0.097	–	–	–	–	–	–	–	–	–	–
Phobic disorders treatment	0.681	0.085	–	–	–	–	–	–	0.516	0.165	–	–	–	–	–	–	–	–	–	–
Phosphodiesterase inhibitor	–	–	–	–	–	–	0.523	0.004	–	–	–	–	–	–	–	–	–	–	–	–
Acetylcholine neuromuscular blocking agent	–	–	–	–	–	–	0.505	0.071	–	–	–	–	–	–	–	–	–	–	–	–
Interferon alpha agonist	0.566	0.005	0.507	0.009	–	–	0.533	0.007	–	–	–	–	–	–	–	–	–	–	–	–
Histidine kinase inhibitor	–	–	0.594	0.016	0.545	0.022	–	–	–	–	0.631	0.013	0.580	0.018					0.584	0.017
Gluconate 2-dehydrogenase (acceptor) inhibitor	–	–	0.505	0.197	0.576	0.140	0.610	0.115	–	–	–	–	0.541	0.167	0.576	0.140	–	–	–	–
Aspulvinone dimethylallyl-transferase inhibitor	–	–	–	–	–	–	–	–	–	–	–	–	0.566	0.110	0.540	0.120	–	–	–	–
Chlordecone reductase inhibitor	–	–	–	–	–	–	–	–	–	–	–	–	0.502	0.083	–	–	–	–	–	–
Antineurotic	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–

Continuation of the Table 1

Prediction of the biological activity spectrum of 1-Ar-4-R-[1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-ones **5***{11-20}* [18]

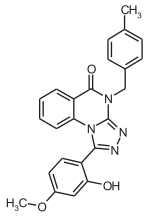
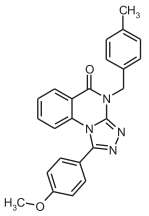
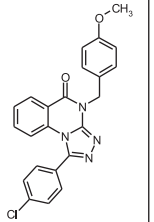
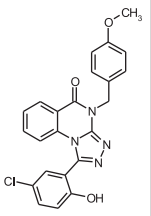
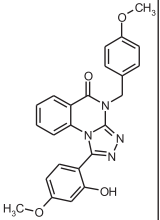
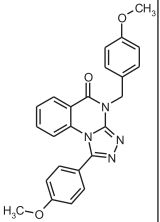
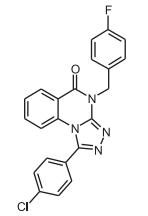
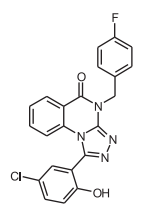
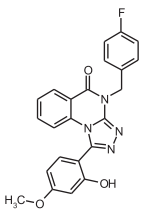
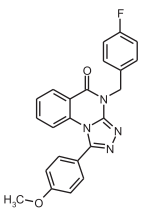
Biological activity	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi	Pa	Pi
	5*{11}*		**5***{12}*		**5***{13}*		**5***{14}*		**5***{15}*		**5***{16}*		**5***{17}*		**5***{18}*		**5***{19}*		**5***{20}*	
																				
Anti-asthmatic	0.599	0.016	0.652	0.011	0.663	0.011	0.606	0.015	0.643	0.012	0.696	0.009	0.647	0.012	0.581	0.017	0.607	0.015	0.658	0.011
Anti-allergic	0.603	0.015	0.632	0.013	0.641	0.012	0.608	0.015	0.636	0.013	0.662	0.011	0.616	0.014	0.580	0.017	0.605	0.015	0.634	0.013
Tumour necrosis factor alpha release inhibitor	–	–	0.572	0.005	0.548	0.006	–	–	0.532	0.532	0.626	0.005	0.576	0.005	–	–	0.534	0.007	0.619	0.005
CYP2A8 substrate	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Tetrahydroxynaphthalene reductase inhibitor	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
5-O-(4-coumaroyl)-D-quinate 3′-monooxygenase inhibitor	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Glycosylphosphatidylinositol phospholipase D inhibitor	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Hepatic function stimulant	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Phobic disorders treatment	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Phosphodiesterase inhibitor	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Acetylcholine neuromuscular blocking agent	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Interferon alpha agonist	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Histidine kinase inhibitor	0.533	0.023	–	–	–	–	0.563	0.020	0.580	0.018	–	–	–	–	0.577	0.018	0.526	0.024	–	–
Gluconate 2-dehydrogenase (acceptor) inhibitor	–	–	0.522	0.182	0.661	0.080	0.630	0.101	0.541	0.167	0.576	0.140	–	–	–	–	–	–	–	–
Aspulvinone dimethylallyl-transferase inhibitor	–	–	–	–	–	–	–	–	0.566	0.110	0.540	0.120	–	–	–	–	–	–	–	–
Chlordecone reductase inhibitor	–	–	–	–	–	–	0.534	0.076	0.502	0.083	–	–	–	–	–	–	–	–	–	–
Antineurotic	–	–	–	–	0.511	0.106	–	–	–	–	–	–	0.551	0.089	–	–	–	–	0.551	0.089

Table 2

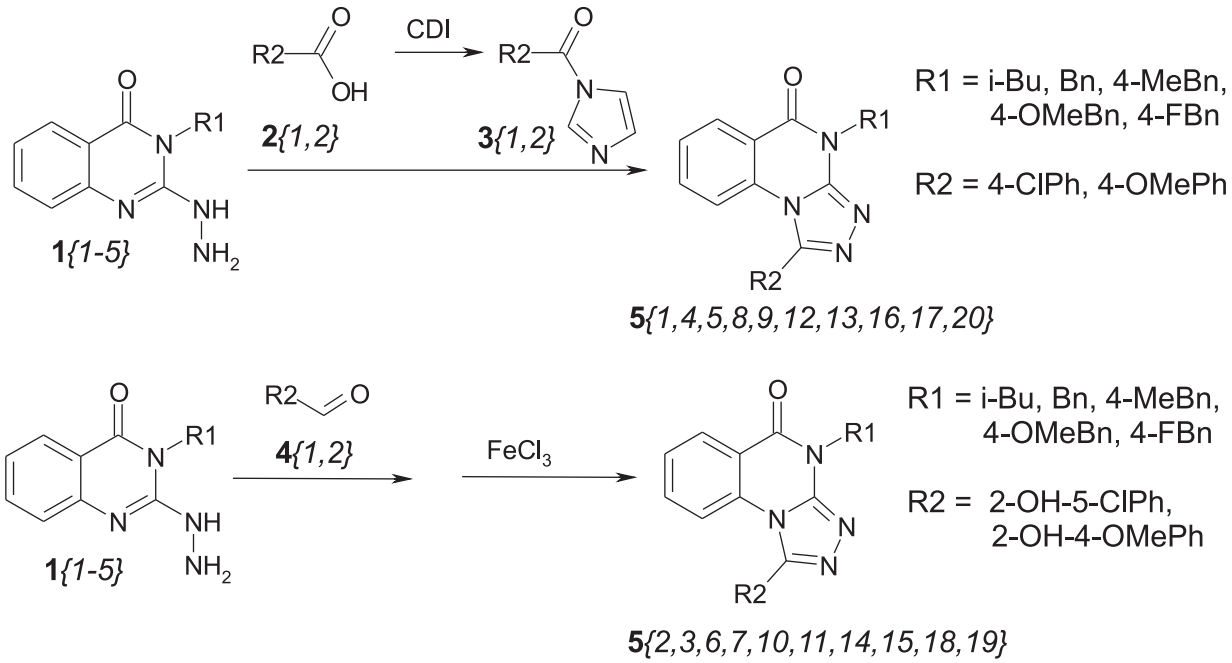
The values of acute toxicity of 1-Ar-4-R-[1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-ones **5***{1-20}* according to the research results **in silico** studied by the GUSAR software [19]

Compound code	LD_{50}, mg/kg			
	Intraperitoneal administration	Intravenous administration	Oral administration	Subcutaneous administration
5*{1}*	493	130	634	1305
5*{2}*	631	112	956	1330
5*{3}*	524	125	1560	935
5*{4}*	391	91	1891	405
5*{5}*	424	181	1227	2595
5*{6}*	790	194	1155	2934
5*{7}*	614	251	1655	2314
5*{8}*	548	182	1124	1284
5*{9}*	633	164	556	2373
5*{10}*	960	165	1024	2335
5*{11}*	564	183	1189	2146
5*{12}*	643	159	1538	1111
5*{13}*	607	165	597	713.7
5*{14}*	679	190	734	1109
5*{15}*	643	222	1172	2354
5*{16}*	410	150	1010	1515
5*{17}*	609	161	976.4	1736
5*{18}*	694	194	851	2225
5*{19}*	450	239	702	707
5*{20}*	482	158	972	560

lowed to eliminate unpromising substances at the early stages of the research.

Prediction of the acute toxicity of compounds **5**{1-20} for different routes of administration (intraperitoneal, intravenous, oral, subcutaneous) was carried out by the GUSAR software [19, 26].

The training set of the programme was developed based on SYMYX MDL Toxicity Database contai-



Scheme

Table 3

Properties of 1-Ar-4-R-[1,2,4]triazolo[4,3-*a*]quinazolin-5(4H)-ones **5***{1-20}*

Compound code	R	Yield, %	M.p., °C	Molecular formula, m.m	N, % Calc./ found	^{1}H NMR spectral data δ, ppm, J, hz
1	2	3	4	5	6	7
5*{1}*	R1 = i-Bu R2 = 4-ClPh	87	246-248	$C_{19}H_{17}ClN_4O$ 352.82	15.88/ 15.90	0.96 d (J 7.0, 6H, $2CH_3$); 2.25-2.40 m (1H, CH); 4.08 d (J 7.0, 2H, CH_2); 7.07 d (J 7.8, 1H, H-9); 7.50 t (J 7.8, 1H, H-7); 7.58-7.78 m (5H, H-8, 2,3,5,6 Ar); 8.23 d (J 7.8, 1H, H-6)
5*{2}*	R1 = i-Bu R2 = 2-OH-5-ClPh	68	227-229	$C_{19}H_{17}ClN_4O_2$ 368.82	15.19/ 15.17	0.96 d (J 7.0, 6H, $2CH_3$); 2.25-2.40 m (1H, CH); 4.08 d (J 7.0, 2H, CH_2); 7.06 d (J 7.8, 1H, H-9); 7.18 d (J 7.8, 1H, H-3 Ar); 7.44-7.60 m (3H, H-7, 4,6 Ar); 7.72 t (J 7.8, 1H, H-8); 8.22 d (J 7.8, 1H, H-6); 10.50 c (1H, OH)
5*{3}*	R1 = i-Bu R2 = 2-OH-4-OMePh	64	214-215	$C_{20}H_{20}N_4O_3$ 364.40	15.38/ 15.40	0.96 d (J 7.0, 6H, $2CH_3$); 2.25-2.40 m (1H, CH); 3.80 c (3H, OCH_3); 4.08 d (J 7.0, 2H, CH_2); 6.54-6.68 m (2H, H-3,5 Ar); 7.22 d (J 7.8, 1H, H-9); 7.36 d (J 7.8, 1H, H-6 Ar); 7.50 t (J 7.8, 1H, H-7); 7.70 t (J 7.8, 1H, H-8); 8.22 d (J 7.8, 1H, H-6); 10.15 c (1H, OH)
5*{4}*	R1 = i-Bu R2 = 4-OMePh	81	233-234	$C_{20}H_{20}N_4O_2$ 348.40	16.08/ 16.05	0.96 d (J 7.0, 6H, $2CH_3$); 2.25-2.40 m (1H, CH); 3.86 c (3H, OCH_3); 4.08 d (J 7.0, 2H, CH_2); 7.05-7.26 m (3H, H-9, 3,5 Ar); 7.44-7.76 m (4H, H-7,8, 2,6 Ar); 8.22 d (J 7.8, 1H, H-6)
5*{5}*	R1 = Bn R2 = 4-ClPh	88	296-298	$C_{22}H_{15}ClN_4O$ 386.83	14.48/ 14.45	5.43 c (2H, CH_2); 7.07 d (J 7.8, 1H, H-9); 7.22-7.36 m (3H, H-3,4,5 Bn); 7.44-7.56 m (3H, H-7, 2,6 Bn); 7.60-7.78 m (5H, H-8, 2,3,5,6 Ar); 8.25 d (J 7.8, 1H, H-6)
5*{6}*	R1 = Bn R2 = 2-OH-5-ClPh	66	278-280	$C_{22}H_{15}ClN_4O_2$ 412.83	13.91/ 13.88	5.43 c (2H, CH_2); 7.11 d (J 7.8, 1H, H-9); 7.17 d (J 7.8, 1H, H-3 Ar); 7.24-7.36 m (3H, H-3,4,5 Bn); 7.44-7.58 m (5H, H-7, 4,6 Ar, 2,6 Bn); 7,81 t (J 7.8, 1H, H-8); 8.26 d (J 7.8, 1H, H-6); 10.50 c (1H, OH)
5*{7}*	R1 = Bn R2 = 2-OH-4-OMePh	62	269-271	$C_{23}H_{18}N_4O_3$ 398.41	14.06/ 14.03	3.80 c (3H, OCH_3); 5.43 c (2H, CH_2); 6,54-6,68 m (2H, H-3,5 Ar); 7.20-7.38 m (5H, H-9, 6 Ar, 3,4,5 Bn); 7.44-7.56 m (3H, H-7, 2,6 Bn); 7.72 t (J 7.8, 1H, H-8); 8.25 d (J 7.8, 1H, H-6); 10.17 c (1H, OH)
5*{8}*	R1 = Bn R2 = 4-OMePh	85	282-284	$C_{23}H_{18}N_4O_2$ 382.41	14.65/ 14.68	3.86 c (3H, OCH_3); 5.43 c (2H, CH_2); 7.07-7.21 m (3H, H-9, 3,5 Ar); 7.24-7.38 m (3H, H-3,4,5 Bn); 7.43-7.56 m (3H, H-7, 2,6 Bn); 7.56-7.72 m (3H, H-8, 2,6 Ar); 8.25 d (J 7.8, 1H, H-6)
5*{9}*	R1 = 4-MeBn R2 = 4-ClPh	84	288-290	$C_{23}H_{17}ClN_4O$ 400.86	13.98/ 14.01	2.24 c (3H, CH_3); 5.38 c (2H, CH_2); 7.03-7.17 m (3H, H-9, 3,5 Bn); 7.38 d (J 7.8, 2H, H-2,6 Bn); 7.52 t (J 7.8, 1H, H-7); 7.62-7.78 m (5H, H-8, 2,3,5,6 Ar); 8.25 d (J 7.8, 1H, H-6)
5*{10}*	R1 = 4-MeBn R2 = 2-OH-5-ClPh	63	268-270	$C_{23}H_{17}ClN_4O_2$ 416.86	13.44/ 13.47	2.24 c (3H, CH_3); 5.38 c (2H, CH_2); 7.00-7.22 m (4H, H-9, 3 Ar, 3,5 Bn); 7.38 d (J 7.8, 2H, H-2,6 Bn); 7.47-7.62 m (3H, H-7, 4,6 Ar); 7.72 t (J 7.8, 1H, H-8); 8.26 d (J 7.8, 1H, H-6); 10.50 c (1H, OH)
5*{11}*	R1 = 4-MeBn R2 = 2-OH-4-OMePh	62	262-264	$C_{24}H_{20}N_4O_3$ 412.44	13.58/ 13.60	2.24 c (3H, CH_3); 3.80 c (3H, OCH_3); 5.38 c (2H, CH_2); 6,54-6,68 m (2H, H-3,5 Ar); 7.12 d (J 7.8, 2H, H-3,5 Bn); 7.22 d (J 7.8, 1H, H-9); 7.32-7.45 m (3H, H-6 Ar, 2,6 Bn); 7.50 t (J 7.8, 1H, H-7); 7.72 t (J 7.8, 1H, H-8); 8.25 d (J 7.8, 1H, H-6); 10.17 c (1H, OH)
5*{12}*	R1 = 4-MeBn R2 = 4-OMePh	85	282-283	$C_{24}H_{20}N_4O_2$ 396.44	14.13/ 14.16	2.24 c (3H, CH_3); 3.86 c (3H, OCH_3); 5.38 c (2H, CH_2); 7.07-7.21 m (5H, H-9, 3,5 Ar, 3,5 Bn); 7.38 d (J 7.8, 2H, H-2,6 Bn); 7.50 t (J 7.8, 1H, H-7); 7.56-7.70 m (3H, H-8, 2,6 Ar); 8.25 d (J 7.8, 1H, H-6)
5*{13}*	R1 = 4-OMeBn R2 = 4-ClPh	88	>300	$C_{23}H_{17}ClN_4O_2$ 416.86	13.44/ 13.42	3.70 c (3H, OCH_3); 5.34 c (2H, CH_2); 6.87 d (J 7.8, 2H, H-3,5 Bn); 7.06 d (J 7.8, 1H, H-9); 7.42-7.54 m (3H, H-7, 2,6 Bn); 7.58-7.78 m (5H, H-8, 2,3,5,6 Ar); 8.24 d (J 7.8, 1H, H-6)
5*{14}*	R1 = 4-OMeBn R2 = 2-OH-5-ClPh	72	294-296	$C_{23}H_{17}ClN_4O_3$ 432.86	12.94/ 12.93	3.70 c (3H, OCH_3); 5.35 c (2H, CH_2); 6.87 d (J 7.8, 2H, H-3,5 Bn); 7.05 d (J 7.8, 1H, H-9); 7.16 d (J 7.8, 1H, H-3 Ar); 7.42-7.58 m (5H, H-7, 4,6 Ar, 2,6 Bn); 7.72 t (J 7.8, 1H, H-8); 8.24 d (J 7.8, 1H, H-6); 10.50 c (1H, OH)
5*{15}*	R1 = 4-OMeBn R2 = 2-OH-4-OMePh	70	288-289	$C_{24}H_{20}N_4O_4$ 428.44	13.08/ 13.11	3.70 c (3H, OCH_3-Bn); 3.80 c (3H, OCH_3-Ar); 5.33 c (2H, CH_2); 6.54-6.68 m (2H, H-3,5 Ar); 6.87 d (J 7.8, 2H, H-3,5 Bn); 7.22 d (J 7.8, 1H, H-9); 7.34 d (J 7.8, 1H, H-6 Ar); 7.42-7.58 m (3H, H-7, 2,6 Bn); 7.72 t (J 7.8, 1H, H-8); 8.24 d (J 7.8, 1H, H-6); 10.48 c (1H, OH)
5*{16}*	R1 = 4-OMeBn R2 = 4-OMePh	86	>300	$C_{24}H_{20}N_4O_3$ 412.44	13.58/ 13.61	3.70 c (3H, OCH_3-Bn); 3.88 c (3H, OCH_3-Ar); 5.37 c (2H, CH_2); 6.87 d (J 7.8, 2H, H-3,5 Bn); 7.15-7.26 m (3H, H-9, 3,5 Ar); 7.44-7.54 m (3H, H-7, 2,6 Bn); 7.58-7.76 m (3H, H-8, 2,6 Ar); 8.24 d (J 7.8, 1H, H-6)

Continuation of the Table 3

1	2	3	4	5	6	7
5*{17}*	R1 = 4-FBn R2 = 4-ClPh	91	>300	$C_{22}H_{14}ClFN_4O$ 404.82	13.84/ 13.82	5.43 c (2H, CH_2); 7.04-7.22 m (3H, H-9, 3,5 Bn); 7.48-7.60 m (3H, H-7, 2,6 Bn); 7.62-7.78 m (5H, H-8, 2,3,5,6 Ar); 8.25 d (J 7.8, 1H, H-6)
5*{18}*	R1 = 4-FBn R2 = 2-OH-5-ClPh	70	>300	$C_{22}H_{14}ClFN_4O_2$ 420.82	13.31/ 13.28	5.43 c (2H, CH_2); 7.08 d (J 7.8, 1H, H-9); 7.14-7.21 m (3H, H-3 Ar, 3,5 Bn); 7.44-7.62 m (5H, H-7, 4,6 Ar, 2,6 Bn); 7.73 t (J 7.8, 1H, H-8); 8.26 d (J 7.8, 1H, H-6); 10.44 c (1H, OH)
5*{19}*	R1 = 4-FBn R2 = 2-OH-4-OMePh	66	296-298	$C_{23}H_{17}FN_4O_3$ 416.41	13.45/ 13.48	3.80 c (3H, OCH_3); 5.40 c (2H, CH_2); 6.52-6.68 m (2H, H-3,5 Ar); 7.07-7.26 m (3H, H-9, 3,5 Bn); 7.35 d (J 7.8, 1H, H-6 Ar); 7.48-7.62 m (3H, H-7, 2,6 Bn); 7.72 t (J 7.8, 1H, H-8); 8.24 d (J 7.8, 1H, H-6); 10.17 c (1H, OH)
5*{20}*	R1 = 4-FBn R2 = 4-OMePh	88	>300	$C_{23}H_{17}FN_4O_2$ 400.41	13.99/ 14.02	3.86 c (3H, OCH_3); 5.38 c (2H, CH_2); 7.07-7.22 m (5H, H-9, 3,5 Ar, 3,5 Bn); 7.45-7.74 m (6H, H-7,8, 2,6 Ar, 2,6 Bn); 8.24 d (J 7.8, 1H, H-6)

ning information about the acute toxicity of more than 10000 chemical structures. The baseline information regarding the acute toxicity of the compounds under study are presented by LD_{50} values (log 10 (mmol/kg) and mg/kg) and the toxicity class according to the OECD classification project of chemical substances by acute toxicity values [19, 20].

The ^{1}H NMR-spectra of compounds **5***{1-20}* were recorded on a Varian WXR-400 (200 MHz) spectrometer in DMSO-d6 solution with TMS as an internal standard, chemical shifts were reported in ppm. Melting points were measured with a Buchi B-520 melting point apparatus. Elemental analysis was performed on an Euro EA-3000 apparatus. Starting 2-hydrazinoquinazolin-4(3*H*)-ones **1***{1-5}* were obtained according to the method [21].

The general procedure for the synthesis of 1-Ar-4-R-[1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-ones 5*{1, 4, 5, 8, 9, 12, 13, 16, 17, 20}*. Dissolve the mixture of 1.5 mmol of the corresponding acid **2***{1, 2}* and 0.24 g (1.5 mmol) of carbonyldiimidazole (CDI) in 5 ml of anhydrous DMFA. Heat the solution at 100°C for 1 h, then add 1 mmol of the corresponding 2-hydrazinoquinazolin-4(3*H*)-one **1***{1-5}*. Reflux the reaction mixture for 24 h. After cooling dilute the reaction mixture with 10 ml of water. The next day filter the precipitate formed, wash with 20 ml of *i*-propanol and recrystallize from the mixture of 5 ml of DMFA and 10 ml of *i*-propanol. Yields and ^{1}H NMR data are given in Table 3.

The general procedure for the synthesis of 1-Ar-4-R-[1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-ones 5*{2, 3, 6, 7, 10, 11, 14, 15, 18, 19}*. Heat the solution of 1 mmol of the corresponding 2-hydrazinoquinazolin-4(3*H*)-one **1***{1-5}* and 1 mmol of the corresponding salicylic aldehyde **4***{1, 2}* in 5 ml of anhydrous DMFA at 100°C for 2 h, then add the solution of 0.54 g (2 mmol) $FeCl_3 \cdot 6\ H_2O$ in 5 ml of DMFA. Heat the reaction mixture at 130°C for 4 hs. After cooling dilute the reaction mixture with 10 ml of water. The next day filter the precipitate formed, wash with 20 ml of *i*-propanol and recrystallize from the mixture of 5 ml of DMFA and 10 ml of *i*-propanol. Yields and ^{1}H NMR data are given in Table 3.

Conclusions

According to the result of computer prediction of the biological activity spectrum and the acute toxicity of 1-Ar-4-R-[1,2,4]triazolo[4,3-*a*]quinazolin-5(4*H*)-ones the selection of slightly toxic or nontoxic substances with the potential anti-asthmatic and anti-allergic activity has been performed.

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THE ALKYLATION REACTION OF AROMATIC ACIDSHYDRAZIDES WITH (±)-*CIS*-3-DICHLOROMETHYL-1,2,2-TRIMETHYLCYCLOPENTANCARBOXYLIC ACID

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Key words: (±)-cis-3-dichloromethyl-1,2,2-trimethylcyclopentancarboxylic acid; hydrazides; hydrazones; alkylation

The article describes the study of the alkylation reaction of aromatic acids hydrazides with (±)-cis-3-dichloromethyl-1,2,2-trimethylcyclopentancarboxylic acid. The acid mentioned is a new substance obtained by oxidative cleavage of racemic camphor in the tetrachloromethane medium according to the method described earlier and modified by us. As a result of alkylation of aromatic acids hydrazides, a series of 3-{[2-(R-carbonyl)hydrazinylidene]methyl}-1,2,2-trimethylcyclopentancarboxylic acids has been obtained with the yields of 77-88%. According to the data of 1H NMR spectra almost all products are E-isomers. The reaction of alkylation of anthranilic hydrazide proceeds with formation of the 1,2,3,4-tetrahydroquinazolin-4-one cycle and obtaining of (±)-cis-3-(3-amino-1,2,3,4-tetrahydroquinazolin-4-on-2-yl)-1,2,2-trimethylcyclopentancarboxylic acid. The composition of the compounds synthesized has been proven by elemental analysis , and their structure has been confirmed by ^{1}H NMR spectroscopy. According to the results of PASS prediction the compounds synthesized are potential diuretic, antiviral and antibacterial agents. The synthetic studies conducted show the possibility of using (±)-cis-3-dichloromethyl-1,2,2-trimethylcyclopentancarboxylic acid as a building block for extension of a number of biologically active substances synthesized in our previous studies on the basis of (±)-cis-1,2,2-trimethylcyclopentan-1,3-dicarboxylic (camphoric) acid.

ДОСЛІДЖЕННЯ РЕАКЦІЇ АЛКІЛУВАННЯ ГІДРАЗИДІВ АРОМАТИЧНИХ КИСЛОТ (±)-ЦИС-3-ДИХЛОРОМЕТИЛ-1,2,2-ТРИМЕТИЛЦИКЛОПЕНТАНКАРБОНОВОЮ КИСЛОТОЮ

Є.О.Цапко

Ключові слова: *(±)-цис-3-дихлорометил-1,2,2-триметилциклопентанкарбонова кислота; гідразиди; гідразони; алкілування*

Роботу присвячено дослідженню реакції алкілування гідразидів ароматичних кислот (±)-цис-3-дихлорометил-1,2,2-триметилциклопентанкарбоновою кислотою. Зазначена кислота є новою сполукою, яку ми отримали окисним розщепленням рацемічної камфори у середовищі тетрахлорометану за методикою, описаною в літературі і модифікованою нами. У результаті алкілування гідразидів ароматичних кислот нами отримано ряд (±)-цис-3-{[2-(R-карбоніл)гідразиніліден]метил}-1,2,2-триметилциклопентанкарбонових кислот з виходом 77-88%. За даними спектрів ^{1}H ЯМР встановлено, що практично всі продукти є Е-ізомерами. Реакція алкілування антранілового гідразиду перебігає із замиканням 1,2,3,4-тетрагідрохіназолін-4-онового циклу та утворенням (±)-цис-3-(3-аміно-1,2,3,4-тетрагідрохіназолін-4-он-2-іл)-1,2,2-триметилциклопентанкарбонової кислоти. Склад синтезованих речовин доведений елементним аналізом, а будова підтверджена методом ^{1}H ЯМР-спектроскопії. За результатами віртуального прогнозу PASS синтезовані сполуки є потенційними діуретичними, антивірусними та антибактеріальними засобами. Проведені синтетичні дослідження показують можливість використання (±)-цис-3-дихлорометил-1,2,2-триметилциклопентанкарбонової кислоти як білдінг блоку для розширення ряду біологічно активних речовин, синтезованих нами у попередніх дослідженнях на основі (±)-цис-1,2,2-триметилциклопентан-1,3-дикарбонової (камфорної) кислоти.

ИССЛЕДОВАНИЕ РЕАКЦИИ АЛКИЛИРОВАНИЯ ГИДРАЗИДОВ АРОМАТИЧЕСКИХ КИСЛОТ (±)-ЦИС-3-ДИХЛОРМЕТИЛ-1,2,2-ТРИМЕТИЛЦИКЛОПЕНТАНКАРБОНОВОЙ КИСЛОТОЙ

Е.А.Цапко

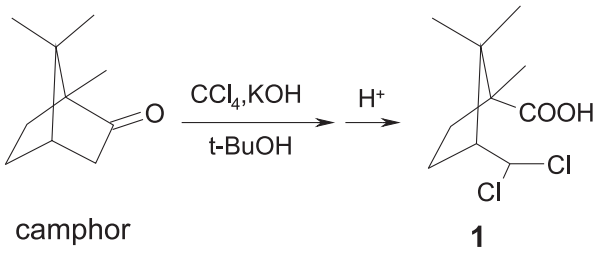
Ключевые слова: *(±)-цис-3-дихлорметил-1,2,2-триметилциклопентанкарбоновая кислота; гидразиды; гидразоны; алкилирование*

Работа посвящена исследованию реакции алкилирования гидразидов ароматических кислот (±)-цис-3-дихлорметил-1,2,2-триметилциклопентанкарбоновой кислотой. Указанная кислота является новым соединением, которое мы получили окислительным расщеплением рацемической камфоры в среде тетрахлорметана по методике, описанной в литературе и модифицированной нами. В результате алкилирования гидразидов ароматических кислот нами получен ряд (±)-цис-3-{[2-(R-карбонил)гидразинилиден]метил}-1,2,2-триметилциклопентанкарбоновых кислот с выходом 77-88%. По данным спектров ^{1}H ЯМР установлено, что практически все продукты являются Е-изомерами. Реакция алкилирования антранилового гидразида протекает с замыканием 1,2,3,4-тетрагидрохиназолин-4-онового цикла и образованием (±)-цис-3-(3-амино-1,2,3,4-тетрагидрохиназолин-4-он-2-ил)-1,2,2-триметилциклопентанкарбоновой кислоты. Состав синтезированных веществ доказан элементным анализом, а строение подтверждено методом ^{1}H ЯМР-спектроскопии. По результатам виртуального прогноза PASS синтезированные соединения являются потенциальными диуретическими, антивирусными и антибактериальными средствами. Проведенные синтетические исследования показывают возможность использования (±)-цис-3-дихлорметил-1,2,2-триметилциклопентанкарбоновой кислоты в качестве билдинг блока для расширения ряда биологически активных веществ, синтезированных нами в предыдущих исследованиях на основе (±)-цис-1,2,2-триметилциклопентан-1,3-дикарбоновой (камфорной) кислоты.

In previous studies we identified derivatives of (±)-*cis*-1,2,2-trimethylcyclopentan-1,3-dicarboxylic (camphoric acid) with the hypoglycemic [1], anticonvulsant [2], diuretic [3] activity. In order to develop new approaches to the synthesis of this series we have obtained 3-dichloromethyl-1,2,2-trimethylcyclopentancarboxylic acid (compound **1**, Scheme 1). Acid **1** and camphoric acid contain the same moiety of 1,2,2-trimethylcyclopentancarboxylic acid.

Acid **1** was obtained by oxidative cleavage of camphor under the action of alkali in the presence of *tert*-butanol in the carbon tetrachloride medium (Scheme 1). For the first time the synthesis of *d*-isomer of acid **1** from *d*-camphor was described by Meyers et al. [4, 5], but the spectral characteristics of the acid were not given, and only chlorolactone was described among its derivatives. We have reproduced and optimized this method for racemic camphor. When carrying out the synthesis we excluded the stages of the solvent stripping and extraction of the product with diethyl ether, as well as reduced the number of *tert*-butanol used. As a result, (±)-acid **1** with the yield of 70% has been obtained, and it coincides with the results of authors [5].

Compound **1** is a white crystalline substance that dissolves in aqueous solutions of alkali and most organic solvents. In ^{1}H NMR spectrum of acid **1** the characteristic group of signals of the 1,2,2-trimethylcyclopentan fragment is present, the signal of the dichloromethyl group proton is observed as a doublet at 6.19 ppm. The mass spectrum of this compound does not have signals of a molecular ion. The heaviest ion has m/z – 167. It is probably formed by cleavage of HCl with the lactone cycle closure and the subsequent cleavage of another chlorine atom.



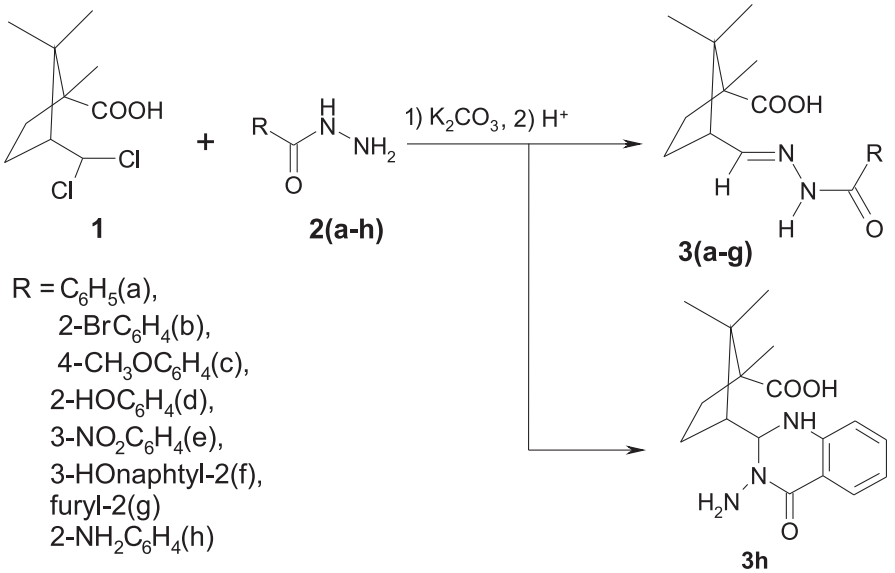
camphor **1**

Scheme 1

There are almost no data published on the use of dichloromethyl aliphatic derivatives in the preparative organic synthesis. This fact can be explained by insufficient accessibility and low reactivity of these derivatives. Only the compounds with an activated dichloromethyl group, such as dichloromethyl ether [6] and dichloromethylphenylsulfoxide [7], were used.

The study of the dichloromethyl group reactivity of compound **1** was started from the alkylation reaction of aromatic acids hydrazides **2** (Scheme 2). The reaction was carried out in the aqueous-alcoholic solution in the presence of potassium carbonate. As a result, a series of 3-{[2-(R-carbonyl)hydrazinylidene] methyl}-1,2,2-trimethylcyclopentancarboxylic acids **3a-g** (Scheme 2) with preparative yields was obtained. All acids **3a-g** obtained are colourless crystalline compounds that are soluble in most organic solvents and practically insoluble in water. Proton signals of the expected structural fragments with the corresponding intensity and multiplicity were observed in ^{1}H NMR spectra of acids **3a-g**, the chemical shift of the azometine group proton was about 7.7 ppm. The proton signal of the NH-group of compounds **3a-g** was observed as one singlet, indicating that the products consisted of a single geometric isomer. The exception is acid **3b** with the volumetric substituent – bromine in the *ortho*-position to the hydrazide group. The integrated intensity of two singlets of NH protons belonging to E- and Z- isomers of acid **3b** is in the ratio of 2:1.

The geometric configuration of compounds **3a-g** was determined on the example of acid **4c** using the homonuclear Overhauser effect [8]. In Fig. the ^{1}H NMR spectrum of this compound is saturation of the signal of N=CH proton. As it can be seen, the singlet of



R = C_6H_5(a),
2-BrC_6H_4(b),
4-$CH_3OC_6H_4$(c),
2-HOC_6H_4(d),
3-$NO_2C_6H_4$(e),
3-HOnaphtyl-2(f),
furyl-2(g)
2-$NH_2C_6H_4$(h)

Scheme 2

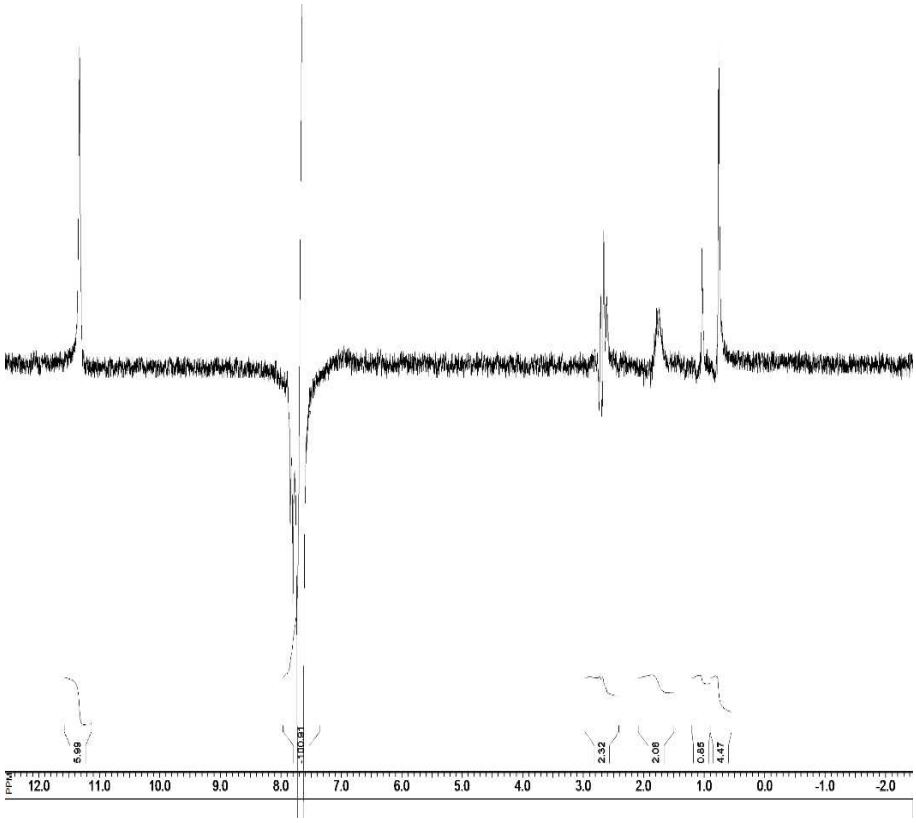


Fig. The ^{1}H NMR spectrum of (±)-*cis*-3-{(E)-2-[(4-methoxybenzoyl)hydrazineylidene]methyl}-1,2,2-trimethylcyclopentanecarboxylic acid 3c with saturation of the signal at 7.76 ppm.

the nearest NH proton (11.32 ppm) is characterized by the biggest integrated intensity indicating E-configuration of the compound studied. Four other signals belong to protons of CH, CH_2 and two CH_3 groups. It is obvious that for compounds **3(a-g)** E-configuration is more favourable than Z-configuration since the cyclopentane fragment together with the carboxyl group can create steric hindrance for Z-configuration.

The result of the reaction between acid **1** and anthranilic hydrazide **2h** was different from products **3a-g** previously discussed (Scheme 2). The product of the reaction was identified as (±)-*cis*-3-(3-amino-1,2,3,4-tetrahydroquinazolin-4-on-2-yl)-1,2,2-trimethylcyclopentancarboxylic acid (compound **3h**). The signals of proton of NH-N=C group were not observed in the ^{1}H NMR spectrum of acid **3h**, while signals of the amino group and the 1,2,3,4-tetrahydroquinazolin-4-one moiety were present.

The PASS virtual screening [9] for compounds **3(a-g)** was conducted. Among the aforementioned types of the biological activity studied for derivatives of camphor acid the diuretic activity is predicted for compounds **3(a-g)** with the index P_a in the range of 0.40-0.55. The anticonvulsant action (P_a = 0.5-0.6) is expected for compound **3h** and its virtual N-acyl derivatives. The cytoprotective and antiviral activities have the highest values of P_a (~0.7) for compounds **3(a-g)**, whereas analysis of the literature data shows that hydrazones have antibacterial [10], antiprotozoal [11] and fungicidal [12, 13] properties.

Experimental Part

According to the homonuclear Overhauser effect experiment the ^{1}H NMR spectra were recorded on a Varian Mercury VX-200 (200 MHz) spectrometer, the solvent was DMSO-d_6 with TMS as an internal standard. The mass spectra were obtained on a Varian 1200L spectrometer in full scanning mode in the range of 35-700 *m/z* and EI at 70 eV. Elemental analysis was performed on an EuroVector EA-3000 microanalyzer. Melting points were determined on a Kofler bench. The purity of the compounds synthesized was confirmed by TLC using Silufol UV_{254} plates and the ethyl acetate – chloroform system (3:2 v/v) as an eluent.

(±)-*Cis*-3-dichloromethyl-1,2,2-trimethylcyclopentancarboxylic acid (1). To the solution of 3 g (0.02 Mol) of camphor in 30 ml of tetrachloromethane add 26 g (0.46 Mol) of a freshly crushed potassium hydroxide. To the obtained mixture add 4 ml of *tert*-butanol and stir. Upon completion of the exothermic reaction stirring was continued for 40 min at 70°C. Cool the mixture and extract potassium salt of acid **1** by three portions of water with 50 ml each. Combine aqueous extracts and acidify them with a dilute sulfuric acid. Filter a white precipitate formed and recrystallize from aqueous ethanol. The yield is 70%. M.p. – 154-155°C. $C_{10}H_{16}Cl_2O_2$. ^{1}H NMR, δ, ppm: 0.80c (3H, CH_3), 1.08c (3H, CH_3), 1.19c (3H, CH_3), 1.23-1.40m, 1.55-1.72m, 1.81-2.02m, 2.14-2.35m (4H, CH_2CH_2), 2.44-2.62m (1H, CH), 6.19d (1H, $CHCl_2$, J = 8 Hz), 12,08s

(1H, COOH). IE-MS, m/z(I_{rel},%): 167(25), 121(100), 116(35), 109(34), 87(56), 83(51), 81(41).

The general procedure for the synthesis of (±)-*cis*-3-([2-(R-carbonyl)hydrazineylidene]methyl)-1,2,2-trimethylcyclopentancarboxylic acids (3). Dissolve 2.4 g (0.01 Mol) of acid **1** and 1.2 g (0.01 Mol) of potassium carbonate in 8 ml of 20% ethanol, add 0.011 Mol of the corresponding hydrazide **2** and heat under the reflux for 2 h. Cool the solution, acidify with a dilute sulfuric acid. Filter the precipitate formed and recrystallize from aqueous ethanol.

(±)-*Cis*-3-{(E)-[(2-benzoyl)hydrazinylidene]methyl}-1,2,2-trimethylcyclopentanecarboxylic acid (3a). The yield is 88%. M.p. – 202-203°C. Found, %: C 67.61, H 7.29, N 9.39. $C_{17}H_{22}N_2O_3$. Calculated, %: C 67.53, H 7.33, N 9.26. ^{1}H NMR, δ, ppm: 0.77c (3H, CH_3), 1.05c (3H, CH_3), 1.15c (3H, CH_3), 1.33-1.51m (1H, CH_2CH_2), 1.68-1.86m (2H, CH_2CH_2), 2.34-2.45m (1H, CH_2CH_2), 2.62-2.79m (1H, CH), 7.40-7.56m (3H, H-3,4,5), 7.69d (1H, CH=N, J=4 Hz), 7.85d (2H, H-2,6, J=4 Hz), 11.44s (1H, NH), 12.16bs (1H, COOH).

(±)-*Cis*-3-{2-[(2-bromobenzoyl)hydrazinylidene]methyl}-1,2,2-trimethylcyclopentanecarboxylic acid (3b). The yield is 87%. M.p. – 244-245°C. Found, %: C 53.44, H 5.59, N 7.46. $C_{17}H_{21}BrN_2O_3$. Calculated, %: C 53.55, H 5.51, N 7.35. ^{1}H NMR, δ, ppm: 0.71c (3H, CH_3), 1.03c (3H, CH_3), 1.15c (3H, CH_3), 1.33-1.47m (1H, CH_2CH_2), 1.56-1.62m (1H, CH_2CH_2), 1.70-1.85m (1H, CH_2CH_2), 2.29-2.43m (1H, CH_2CH_2), 2.63-2.78m (1H, CH), 7.26-7.69m (5H,C_6H_4+CH=N), 11.54s (1H, NH), 12.14bs (1H, COOH).

(±)-*Cis*-3-{(E)-2-[(4-methoxybenzoyl)hydrazinylidene]methyl}-1,2,2-trimethylcyclopentanecarboxylic acid (3c). The yield is 87%. M.p. – 157-158°C. Found, %: C 65.00, H 7.33, N 8,52. $C_{18}H_{24}N_2O_4$. Calculated, %: C 65.04, H 7.28, N 8.43. ^{1}H NMR, δ, ppm: 0.77c (3H, CH_3), 1.03c (3H, CH_3), 1.15c (3H, CH_3), 1.33-1.50м (1H, CH_2CH_2), 1.67-1.85м (2H, CH_2CH_2), 2.34-2.45м (1H, CH_2CH_2), 2.60-2.77м (1H, CH), 3.80s (3H, CH_3O), 7.00d (2H, H-3,5, J=12 Hz), 7.67d (1H, CH=N, J = 8 Hz), 7.82d (2H, H-2,6, J = 12 Hz), 11.32s (1H, NH), 12.14bs (1H, COOH).

(±)-*Cis*-3-{(E)-2-[(2-hydroxybenzoyl)hydrazinylidene]methyl}-1,2,2-trimethylcyclopentanecarboxylic acid (3d). The yield is 77%. M.p. – 172-173°C. Found, %: C 64.15, H 7.03, N 9.01. $C_{17}H_{22}N_2O_4$. Calculated, %: C 64.13, H 6.97, N 8.80. ^{1}H, NMR, δ, ppm: 0.78c (3H, CH_3), 1.06c (3H, CH_3), 1.15c (3H, CH_3), 1.34-1.51m (1H, CH_2CH_2), 1.70-1.89m (2H, CH_2CH_2), 2.34-2.46m (1H, CH_2CH_2), 2.64-2.80m (1H, CH), 6.84-6.96m (2H, H-5+CH=N), 7.40t (1H, H-4, J = 8Hz) 7.72d (1H, H-3, J = 8 Hz), 7.82d (1H, H-6, J = 8 Hz), 9.03s (1H, OH) 11.54s (1H, NH), 12.02bs (1H, COOH).

(±)-*Cis*-3-{(E)-2-[(3-nitrobenzoyl)hydrazinylidene]methyl}-1,2,2-trimethylcyclopentanecarboxylic acid(3e). The yield is 86%. M.p. – 188-189°C. Found, %: C 58.84, H 6.16, N 12.19. $C_{17}H_{21}N_3O_5$. Calculated, %: C 58.78, H 6.09, N 12.10. ^{1}H NMR, δ, ppm: 0.79c (3H, CH_3), 1.05c (3H, CH_3), 1.15c (3H, CH_3), 1.32-1.53m (1H, CH_2CH_2), 1.69-1.92m (2H, CH_2CH_2), 2.30-2.45m (1H, CH_2CH_2), 2.64-2.84m (1H, CH), 7.68-7.88m (2H, H-3+CH=N), 8.29d (1H, H-2, J = 8 Hz), 8.39d (1H, H-4, J = 8 Hz), 8.69s (1H, H-6), 11.78s (1H, NH), 12.16bs (1H, COOH).

(±)-*Cis*-3-{(E)-2-[(3-hydroxynaphthalen-2-ylcarbonyl)hydrazinylidene]methyl}-1,2,2-trimethylcyclopentanecarboxylic acid(3f). The yield is 71%. M.p. – 229-230°C. Found, %: C 68.44, H 6.55, N 7.69. $C_{21}H_{24}N_2O_4$. Calculated, %: C 68.46, H 6.57, N 7.60. ^{1}H NMR, δ, ppm: 0.79c (3H, CH_3), 1.07c (3H, CH_3), 1.17c (3H, CH_3), 1.25-1.50m (2H, CH_2CH_2), 1.74-1.89m (2H, CH_2CH_2), 2.70-2.85m (1H, CH), 7.31-7.87m (7H, CH=N+naphthyl), 8.39s (1H, OH), 11.68s (1H, NH), 12.16bs (1H, COOH).

(±)-*Cis*-3-{2-[(furil-2-ylcarbonyl)hydrazinylidene]methyl}-1,2,2-trimethylcyclopentanecarboxylic acid(3g). The yield is 88%. M.p. – 203-204°C. Found, %: C 61.61, H 6.96, N 9.70. $C_{15}H_{20}N_2O_4$. Calculated, %: C 61.63, H 6.90, N 9.58. ^{1}H NMR, δ, ppm: 0.75c (3H, CH_3), 1.04c (3H, CH_3), 1.14c (3H, CH_3), 1.68-1.90m (2H, CH_2CH_2), 1.29-1.50m, 2.29-2.51m (2H, CH_2CH_2), 2.70-2.85m (1H, CH), 6.62-6.66m (1H, H-4), 7.17-7.22m (1H, H-3), 7.65-7.71m (1H, CH=N), 7.86-7.89m (1H, H-5),11.42s (1H, NH), 12.16bs (1H, COOH).

(±)-*Cis*-3-(3-amino-1,2,3,4-tetrahydroquinazolin-4-on-2-yl)-1,2,2-trimethylcyclopentanecarboxylic acid (3h). The yield is 47%. M.p. – 159-160°C. Found, %: C 64.39, H 7.28, N 13.28. $C_{17}H_{23}N_3O_3$. Calculated, %: C 64.33, H 7.30, N 13.24. ^{1}H NMR, δ, ppm: 0.86c (3H, CH_3), 0.88c (3H, CH_3), 1.01c (3H, CH_3), 1.13-1.32м, 1.48-1.70м, 1.76-1.99м (3H, CH_2CH_2), 2.20-2.43м (2H, C<u>H</u>$_2$C<u>H</u>),4.68dd (1H, NH, J = 8 Hz, 4Hz) 4.86s (2H, NH_2), 6.61-6.71m (2H, H-2,6, J = 8 Hz), 6.93d (1H, H-8, J = 4 Hz), 7.22t (1H, H-7, J = 8 Hz), 7.58d (1H, H-5, J = 8 Hz), 12.16bs (1H, COOH).

Conclusions

1. The synthesis of (±)-*cis*-3-dichloromethyl-1,2,2-trimethylcyclopentancarboxylic acid has been carried out, and its structure has been confirmed by the methods of ^{1}H NMR spectroscopy and mass spectrometry.

2. As a result of the alkylation reaction of aromatic acids hydrazides with (±)-*cis*-3-dichloromethyl-1,2,2-trimethylcyclopentancarboxylic acid a number of the corresponding hydrazones has been obtained. Almost all acids are E-isomers, and it has been confirmed by ^{1}H NMR spectroscopy.

3. It has been found that the alkylation reaction of anthranilic hydrazide with (±)-*cis*-3-dichloromethyl-1,2,2-trimethylcyclopentancarboxylic acid completes with formation of the 1,2,3,4-tetrahydroquinazolin-4-one cycle and obtaining of (±)-*cis*-3-(3-amino-1,2,3,4-tetrahydroquinazolin-4-on-2-yl)-1,2,2-trimethylcyclopentancarboxylic acid.

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THE STUDY OF DEGRADATION PRODUCTS OF CHLORPROMAZINE HYDROCHLORIDE BY THE METHOD OF LIQUID-MASS SPECTROSCOPY IN DRUGS FOR INJECTION

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Key words: chlorpromazine hydrochloride; chlorpromazine sulfoxide; chlorpromazine N-oxide; nor-chlorpromazine; chromatography mass-spectrometry; degradation products

The analysis of model samples of the drug Aminazin has been performed, its active substance is chlorpromazine hydrochloride. These model solutions were subjected to sterilization with previous exposure at room temperature. It has been found that depending on duration of the exposure during sterilization the degradation of the drug occurs in two possible ways – with formation of opalescence or without it. The schemes of degradation with formation of either one product – chlorpromazine sulfoxide, or some products – chlorpromazine sulfoxide, chlorpromazine N-oxide, nor-chlorpromazine and others have been proposed. It has been found that opalescence of solutions is caused by formation of degradation products – chlorpromazine N-oxide and nor-chlorpromazine that are slightly soluble in water. The analysis of model samples was performed using the liquid chromatography/mass spectrometry methods developed by the authors. In the course of analysis the molecular weights corresponding to the abovementioned products were obtained. Therefore, the experimental confirmation of the schemes of degradation proposed has been obtained. As the result of the research conducted the chromatographic method for detection of impurities using liquid chromatography/mass spectrometry has been developed, and the main ways of degradation of chlorpromazine hydrochloride in aqueous solutions have been determined. According to the result of the research the recommendations to the manufacturing process have been developed, and measures for optimizing the composition of the drug have been proposed.

ВИВЧЕННЯ ПРОДУКТІВ ДЕГРАДАЦІЇ ХЛОРОПРОМАЗИНУ ГІДРОХЛОРИДУ МЕТОДОМ РІДИННОЇ МАС-СПЕКТРОСКОПІЇ У ІН'ЄКЦІЙНИХ ПРЕПАРАТАХ
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Ключові слова: *хлоропромазину гідрохлорид; хлоропромазину сульфоксид; хлоропромазину N-оксид; нор-хлоропромазин; хроматомас-спектрометрія; продукти деградації*
Проведений аналіз модельних зразків препарату Аміназин, діючою речовиною якого є хлоропромазину гідрохлорид. Вказані модельні розчини пройшли стерилізацію з попередньою витримкою при кімнатній температурі. Встановлено, що залежно від тривалості експозиції при стерилізації деградація препарату проходить за двома можливими шляхами – з утворенням опалесценції або без неї. Були запропоновані схеми деградації з утворенням або одного продукту хлоропромазину сульфоксиду, або декількох – хлоропромазину сульфоксиду, хлоропромазину N-оксиду, нор-хлопромазину та ін. Було встановлено, що опалесценція розчинів викликається утворенням водонерозчинних продуктів деградації – хлоропромазину N-оксиду, нор-хлопромазину. Аналіз модельних зразків проводився методом рідинної хроматомас-спектрометрії за методикою, розробленою авторами статті. Під час аналізу були отримані молекулярні маси, що відповідають вказаним вище продуктам. Таким чином, було отримано експериментальне підтвердження запропонованих схем деградації. У результаті проведеної роботи нами була розроблена хроматографічна методика виявлення домішок методом рідинної хроматомас-спектрометрії та визначені головні шляхи деградації хлоропромазину гідрохлориду у водних розчинах. Також у результаті проведених досліджень були розроблені рекомендації до виробничого процесу та запропоновані заходи з оптимизації складу препарату.

ИЗУЧЕНИЕ ПРОДУКТОВ ДЕГРАДАЦИИ ХЛОРПРОМАЗИНА ГИДРОХЛОРИДА МЕТОДОМ ЖИДКОСТНОЙ МАСС-СПЕКТРОСКОПИИ В ИНЪЕКЦИОННЫХ ПРЕПАРАТАХ
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Ключевые слова: *хлорпромазина гидрохлорид; хлорпромазина сульфоксид; хлорпромазина N-оксид; нор-хлорпромазин; хроматомасс-спектрометрия; продукты деградации*
Проведен анализ модельных образцов препарата Аминазин, действующим веществом которого является хлорпромазина гидрохлорид. Данные модельные растворы были подвергнуты стерилизации с предварительной выдержкой при комнатной температуре. Установлено, что в зависимости от длительности экспозиции при стерилизации деградация препарата происходит по двум возможным путям – с образованием опалесценции либо без нее. Были предложены схемы деградации с образованием либо одного продукта хлорпромазина сульфоксида, либо нескольких – хлорпромазина сульфоксида, хлорпромазина N-оксида, нор-хлорпромазина и пр. Было установлено, что опалесценция растворов вызывается образованием мало растворимых в воде продуктов деградации –хлорпромазина N-оксида, нор-хлопромазина. Анализ модельных образцов проводился методом жидкостной хроматомасс-спектрометрии по методике, разработанной авторами статьи. В ходе анализа были получены молеку-

лярные массы, соответствующие указанным выше продуктам. Таким образом, было получено экспериментальное подтверждение предложенных схем деградации. В результате проведенной работы нами была разработана хроматографическая методика обнаружения примесей методом жидкостной хроматомасс-спектрометрии и определены основные пути деградации хлорпромазина гидрохлорида в водных растворах. Также в результате проведенных исследований были разработаны рекомендации к производственному процессу и предложены меры по оптимизации состава препарата.

Today phenothiazine derivatives are widely used in the pharmaceutical industry. The application of these substances is stipulated by their neuroleptic, antihistaminic and anti-arrhythmic effect on the human body. Chlorpromazine is the first synthesized neuroleptic (1950), a parent compound of phenothiazine drugs (including trifluoperazine, promazine, fluphenazine, etc.) and the basis for many of antipsychotics and antidepressants. In this regard, drugs with antipsychotic activity, in which chlorpromazine hydrochloride is the active pharmaceutical ingredient, are the most widespread at the pharmaceutical market. One of the representatives of such drugs is Aminazin, solution for injection.

The substance of chlorpromazine hydrochloride is described in the articles of such pharmacopoeias as the European Pharmacopoeia PhEur 8.0 "Chlorpromazine hydrochloride" (07/2012:0475) [1], United States Pharmacopeia USP37-NF32 c.2318 and State Pharmacopoeia of Ukraine (SPhU) 1.0 [3]. The USP37-NF32 has also the article [2] containing requirements for the quality control of solutions for injection. However, even the use of these regulations to control the quality of the raw material and finished products is not a guarantee of quality. There are some reasons for it, in particular because of the quality of the substance and the technological process of preparation of the solution since this substance is highly reactive, and the slightest deviation from the validated technologies of the drug production may lead to the loss of quality of the finished product.

According to the IUPAC classification [5] chlorpromazine hydrochloride (2-chloro-10-[3-(dimethylaminopropyl)]phenothiazine hydrochloride) being a derivative of phenothiazine (10H-dibenzo-[b,e]-1,4-thiazine) belongs to heterocyclic compounds containing the atoms of sulphur and nitrogen in the cycle.

A high reactivity of phenothiazine and chlorpromazine [5] is a consequence of the presence of easily oxidizable sulphur atom in the structure of these substances (Fig. 1).

Under the effect of such strong oxidizers as potassium permanganate, hydrogen peroxide these compounds may form phenothiazine oxide-5 and phenotiazine dioxide-5,5 due to oxidation of the sulphur atom. It is confirmed by the requirements of the USP article on control of the content of chlorpromazine sulfoxide in solutions for injection using the thin-layer chromatography [2].

The tertiary nitrogen in the structure of chlorpromazine hydrochloride can also be oxidized in neutral or acidic media to N-oxide. In addition, for phenothiazine and its derivatives the electrophilic substitution reactions, in which these compounds act as electron donors, are typical [8].

Thus, there is a need of inhibition of the oxidation process of chlorpromazine hydrochloride with atmospheric oxygen when preparing the solution of the drug. The solution to this problem was to introduce the substances – sodium metabisulfite, sodium sulfite absorbing oxygen dissolved in the product in the composition of the drug by interacting with it and to purge the solution with nitrogen to displace oxygen.

However, despite the measures taken to remove oxygen from the solution of the drug there are cases of noncompliance of the drug quality to requirements of the Pharmacopoeia by "Transparency" indicator – opalescence is observed in solutions [4].

Therefore, manufacture of a drug corresponding to the requirements of the Pharmacopoeia requires formation of additional requirements to the technological process and the quality of the substance; it can be done knowing the cause of impurities and their structure. To study the possible degradation products of chlorpromazine hydrochloride the LC-MS/MS method has been developed.

In the conditions given bellow the chromatograms of the full ion current were obtained for the samples of the solution of chlopromazine sterilized immediately after preparation and after exposure for 48 h. The samples were prepared from the substance of one and the same manufacturer.

The chromatograms and mass spectra of the substances detected are given in Figures below.

Experimental Part

To study the possible degradation products of chlorpromazine hydrochloride the model solutions corresponding to the composition of the drug were prepared and placed in glass vials similar to those used in the manufacture of the drug Aminazin. All model

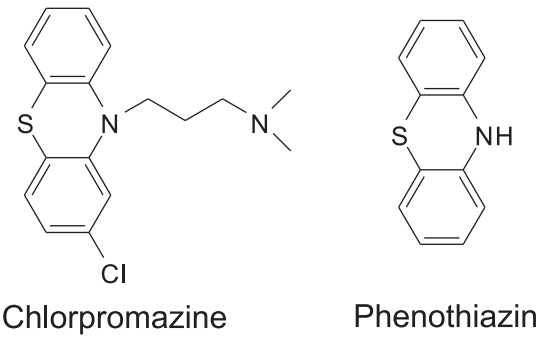

Fig. 1. The structural formulas of phenothiazine and chlorpromazine.

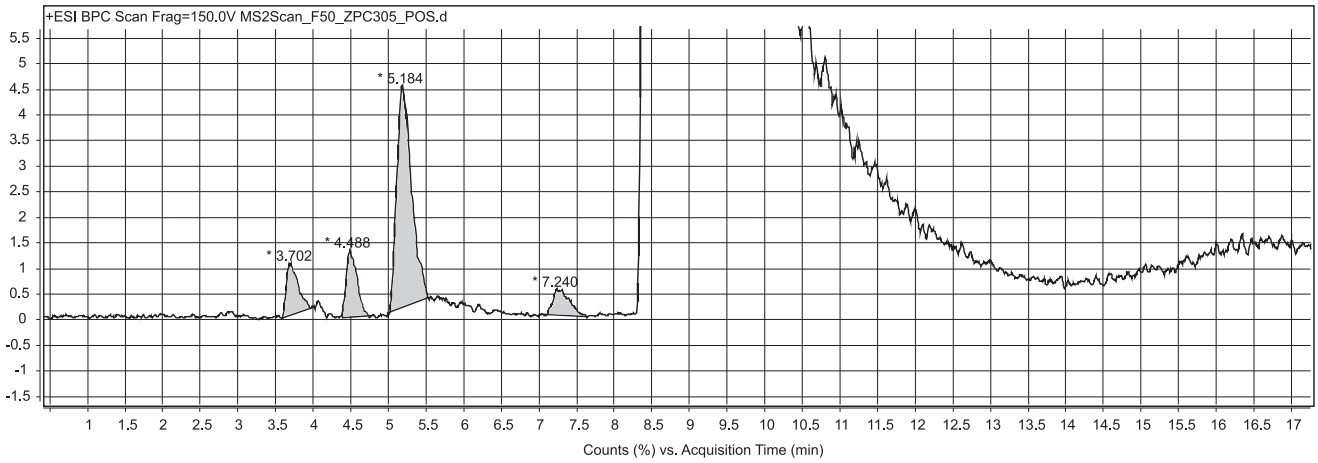

Fig. 2. The chromatogram of the full ion current obtained for the sample immediately after sterilization.

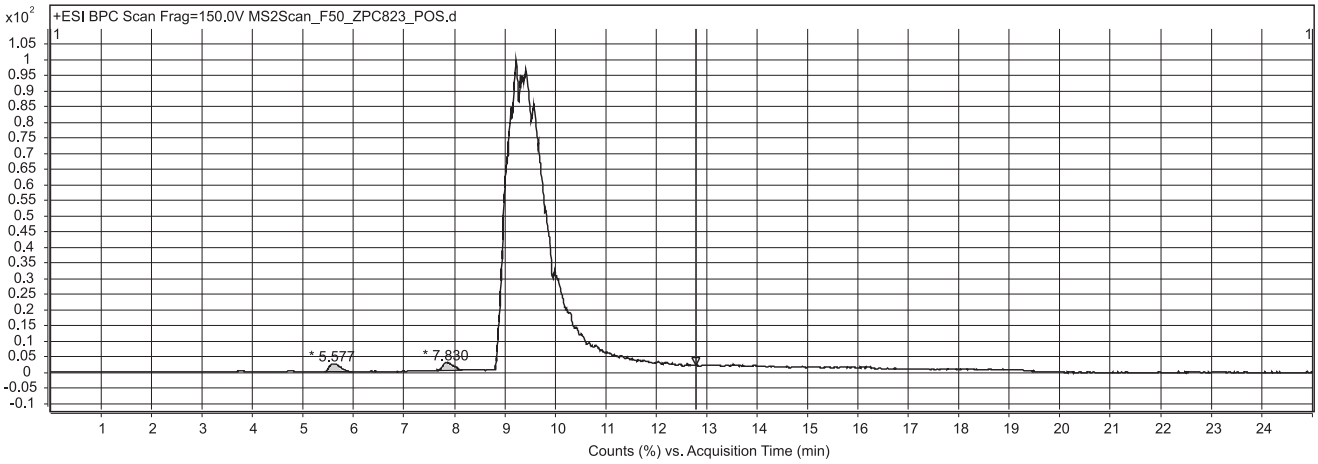

Fig. 3. The chromatogram of the full ion current obtained for the sample sterilized after exposure.

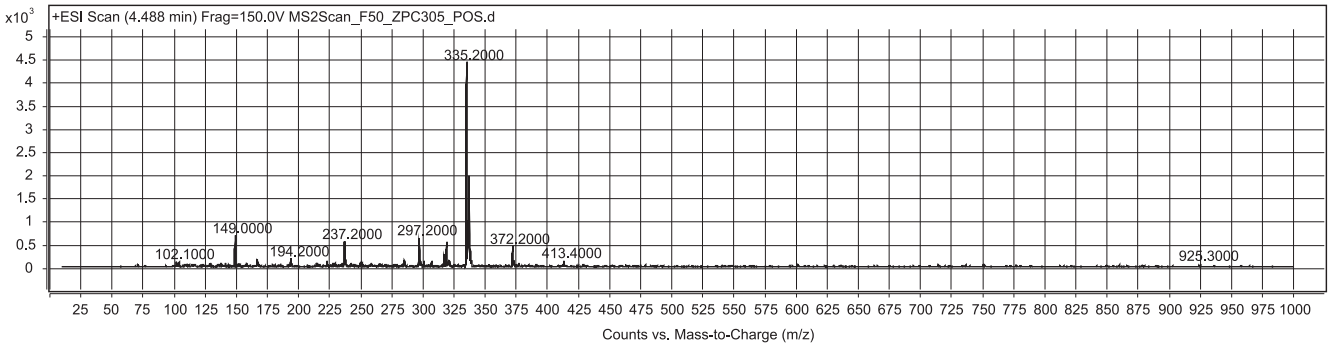

Fig. 4. The mass spectrum of chlorpromazine sulfoxide and N-oxide (the time of escape peak is 5.184 min).

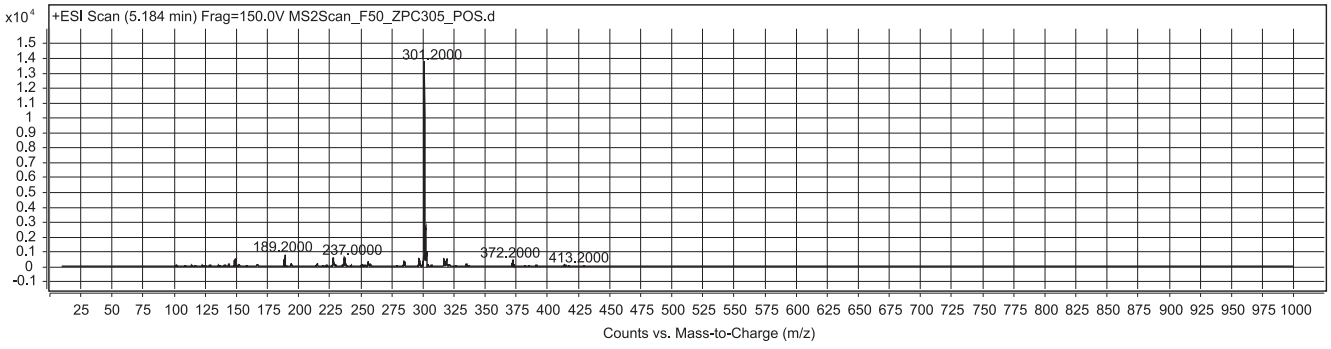

Fig. 5. The mass spectrum of nor-chlorpromazine (the time of escape peak is 4.488 min).

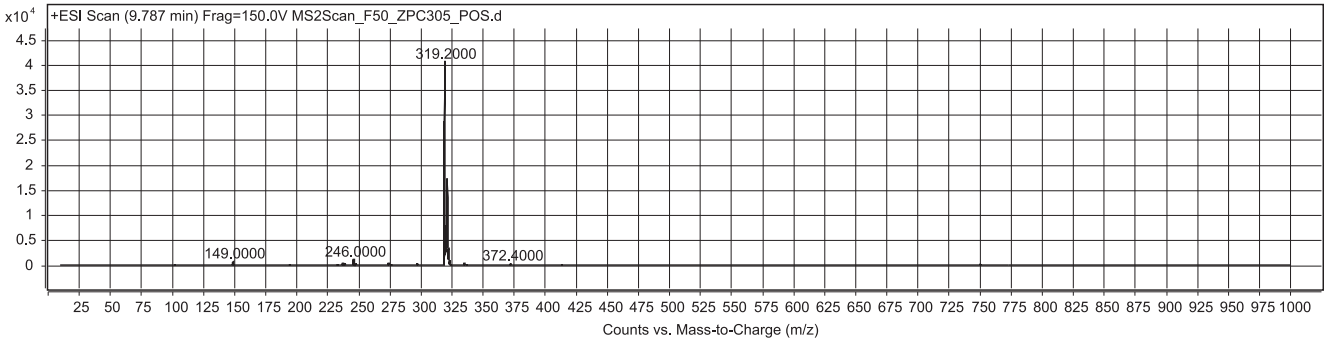

Fig. 6. The mass spectrum of chlorpromazine (the time of escape peak is 9.787 min).

solutions were prepared from the same batch of chlorpromazine hydrochloride substance of the Indian manufacturer. All indicators of the substance quality is fully consistent with the requirements of the article of the European Pharmacopoeia 8.0.

These solutions were sterilized in the conditions corresponding to the conditions of the drug production. It was observed that solutions sterilized immediately after preparation, did not meet the requirements of Pharmacopoeia [2] by "Transparency" indicator – opalescence appeared in these solutions. However, those solutions, in which 48 hours passed from their preparation and sterilization, were fully consistent with the requirements of the article by "Transparency" indicator.

Since for the study of the structure of possible degradation products a highly selective detector is required, an Agilent 6420 Triple Quad mass spectrometer was applied as a detection system. This detector was used together with an Agilent 1100 liquid chromatograph equipped with a diode array, four-channel pump for formation of a low pressure gradient, an autosampler and a column thermostat. To process the measurement results obtained the MassHunter software version B.05.00 was used.

The procedure of conducting chromatographic measurements is given below:

- the column with the size of 250×4.6 mm filled with a sorbent with the bonded phase of octyl silica gel (L1), the particle size is 5 μm, XTerra MS C18 (Waters) or similar, for which the requirements of the section "Chromatographic system suitability" are met;
- the flow rate – 1 ml/min;
- the temperature of the column thermostat – 30°C;
- the mobile phase A: 0.005 M solution of ammonium formate in water *R* degassed in any convenient way;
- the mobile phase B: 0.005 M solution of ammonium formate in the mixture of acetonitrile *R* – water *R* (90:10) degassed in any convenient way;
- the elution mode – gradient.

No.	Time, min	Mobile phase A	Mobile phase B	The elution mode
1	0-5	100	0	Isocratic
2	5-15	100 → 10	0 → 90	Linear gradient
3	15-16	10 → 100	90 → 0	Linear gradient
4	16-20	100	0	Isocratic

- the injection volume – 20 μl;
- detector – mass spectrometer (Agilent 6420 Triple Quad);
- the detector settings:
- the ionization type: positive, electrospray (+ESI);
- the measurement mode: scanning in the mass range – 10-1000 amu;
- the voltage on the fragmentor – 100 V;
- the nitrogen temperature – 350°C;
- the nitrogen consumption – 10 ml/min;
- the nebulizer pressure – 35 PSI;
- the voltage on the capillary – 4 kV.

Results and Discussion

After analyzing the solutions on a liquid chromatography-mass spectrometer (Agilent 6420 Triple Quad) it was found in addition to the main mass of 319 amu the substances with the masses of 335, 305, 285 and 317 $[M+H]^+$ amu were present in solutions. It confirms the presence of chlorpromazine sulfoxide, chlorpromazine N-oxide, nor-chlorpromazine in the solution [6, 7].

Based on the data obtained it was determined that degradation of aqueous solutions of chlorpromazine during sterilization occurred according to the scheme given in Fig. 7. Thus, two possible ways of degradation were considered: with the excess and the lack of oxygen in the drug solution.

The above diagram explains well how in the case of the lack of oxygen (between preparation and sterilization enough time has passed and stabilizers are oxidized removing oxygen from the solution) chlorpromazine is oxidized to chlorpromazine sulfoxide. It is a crystalline substance that is readily soluble in water and does not cause opalescence of the solution. In the case of the oxygen excess (sterilization immediately after preparation, the reducing agents

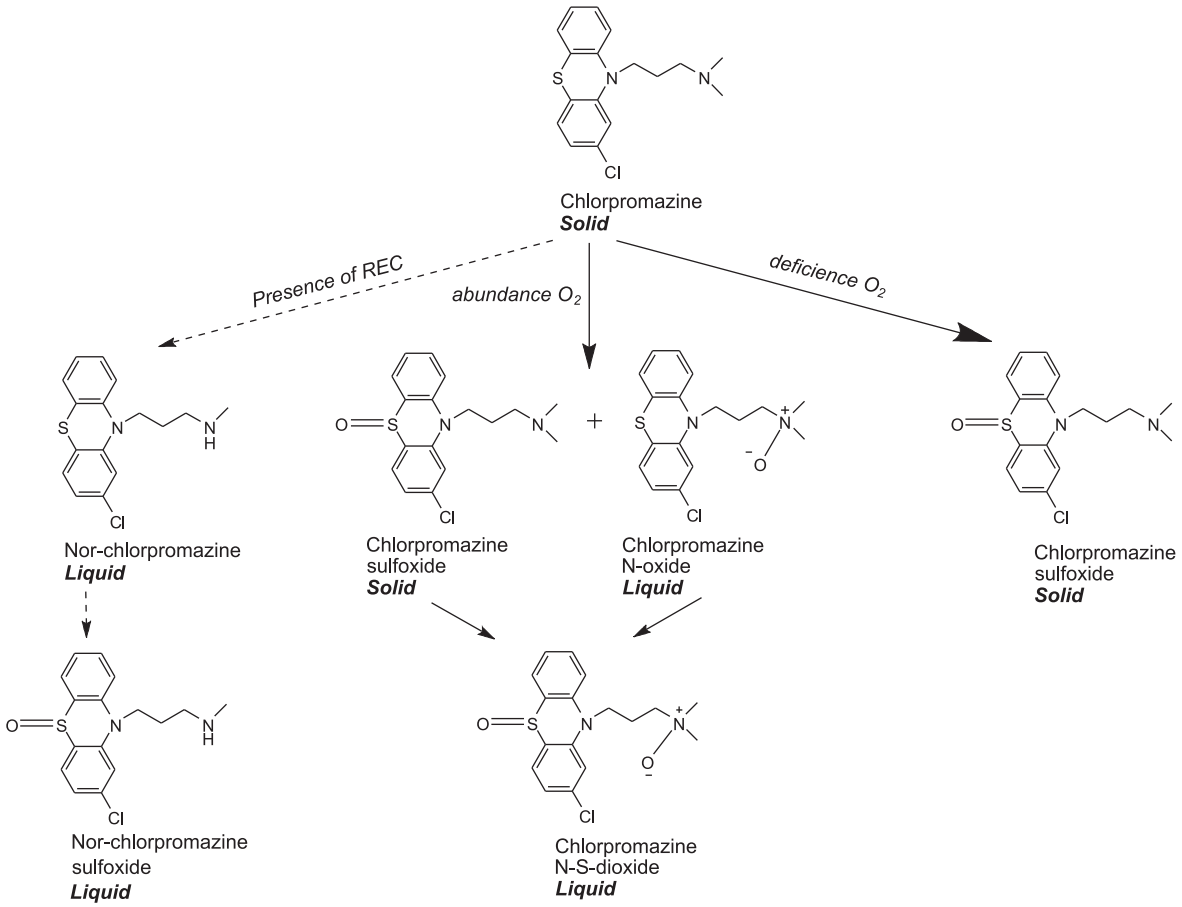

Fig. 7. The ways of degradation of chlorpromazine hydrochloride in an aqueous solution.

in the solution have no time to interact with oxygen) chlorpromazine being oxidized forms simultaneously chlorpromazine sulfoxide and chlorpromazine N-oxide, which can then be oxidized to chlorpromazine N-S-dioxide, and the presence of unused reducing agents in the solution provides a parallel process of formation of nor-chlorpromazine and the corresponding sulfoxide. Since chlorpromazine N-oxide, chlorpromazine N-S-dioxide, nor-chlorpromazine and nor-chlorpromazine sulfoxide are amorphous substances that are poorly soluble in water, the two-phase heterogeneous system is formed in the solution, and it is the cause of opalescence. Impurities of the chlorpromazine substance (for example, chlorphenothiazine) can also be oxidized to sulfoxides; however, since the Pharmacopoeia normalizes the content of impurities in the range of 0.15-0.3%, the contribution of their derivatives to the overall picture of the profile of impurities of the drug is insignificant.

The results obtained are in good agreement with the work of the British scientists [8], in which the principal possibility of formation of the impurities analyzed is shown.

Based on the research results mentioned above one can judge about the nature of possible degradation products; it, in turn, has allowed to develop a number of measures that make possible to obtain the drug, which fully complies with the requirements of Pharmacopoeia. Thus, it has been proposed to perform the exposure of the solution prepared before its sterilization during the manufacturing process, it will allow to remove dissolved oxygen most fully from the solution. It has been also suggested to optimize the process of removing oxygen from the solution of the drug by increasing the purging time and toughening of requirements to the purity of the nitrogen used. However, changes in the technological process is not the only way to provide the quality of the drug prepared. Quite effective way is to optimize the composition of the drug. Therefore, it has been proposed to increase the concentration of substances absorbing oxygen in the composition of the drug, it will reduce the risk of its opalescence in formation of oxidation products of chlorpromazine hydrochloride.

Conclusions

As the result of the research conducted the chromatographic method for detection of impurities using liquid chromatography/mass spectrometry has been developed; the main ways of degradation of chlorpromazine hydrochloride in aqueous solutions have been determined; the recommendations for improving the process of drug manufacture based on the specified substance have been developed, and measures for optimizing the composition of drugs based on chlorpromazine have been proposed.

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THE SYNTHESIS AND PHYSICOCHEMICAL PROPERTIES OF 2-(5-METHOXY-PHENYL-1H-1,2,4-TRIAZOLE-3-YLTHIO) ACETONITRILES AND THEIR IMINOETHERS

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Key words: 1,2,4-triazoles; nitriles; iminoethers

With the purpose of further search of biologically active substances among 5-(2-, 3-, 4-methoxyphenyl, 3,4,5-trimethoxyphenyl)-3-thio-1,2,4-triazoles and their derivatives 10 new compounds have been obtained. Acetonitrilothio-1,2,4-triazoles have been synthesized by alkylation of 5-(2-, 3-, 4-methoxyphenyl and 3,4,5-trimethoxyphenyl)-3-thio-1,2,4-triazoles with halogenonitriles; the primary computer pharmacological screening has shown that the class of compounds mentioned can show such types of the pharmacological activity as antitumor, anti-inflammatory and antioxidant ones. Alkylation of 5-R-1,2,4-triazole-3-thiols has been carried out in the medium of anhydrous alcohol or aprotic solvents. It has been found that replacement of the alcoholic solvent by the aprotic one increases the quantitative yield of 5-R-1,2,4-triazol-3-thioacetonitrile; in aprotic solvents the presence of impurities of alkaline hydrolysis products is not practically observed. Iminoethers of 2-(5-(2-, 3-, 4-methoxyphenyl and 3,4,5-trimethoxyphenyl)-1H-1,2,4-triazole-3-ylthio)acetatic acids have been synthesized by saturation of 2-(5-(2-, 3-, 4-methoxyphenyl and 3,4,5-trimethoxyphenyl)-1H-1,2,4-triazol-3-ylthio)acetonitriles with the flow of dry in the alcoholic medium when constant cooling the reaction mixture to –5ºC. It has been found by the method of HPLC/DAD-MS that the qualitative yield of the target product depends on the maintenance of the temperature mode of the reaction mixture. The structure and individuality of the molecules of the substances synthesized have been proven by the method of ^{1}H NMR-spectroscopy and HPLC/DAD-MS.

СИНТЕЗ І ФІЗИКО-ХІМІЧНІ ВЛАСТИВОСТІ 2-(5-МЕТОКСИФЕНІЛ-1Н-1,2,4-ТРИАЗОЛ-3-ІЛТІО)АЦЕТОНІТРИЛІВ ТА ЇХ ІМІНОЕФІРІВ

Ю.Г.Самелюк, А.Г.Каплаушенко

Ключові слова: *1,2,4-триазоли; нітрили; іміноефіри*

З метою подальшого пошуку біологічно активних речовин серед 5-(2-, 3-, 4-метоксифеніл, 3,4,5-триметоксифеніл)-3-тіо-1,2,4-триазолів та їх похідних було отримано 10 нових сполук. Шляхом алкілування 5-(2-, 3-, 4-метоксифеніл, 3,4,5-триметоксифеніл)-3-тіо-1,2,4-триазолів галогенонітрилами були синтезовані ацетонітрилтіо-1,2,4-триазоли. Первинний комп'ютерний фармакологічний скринінг показав, що цей клас сполук може проявляти такі види фармакологічної активності, як протипухлинна, протизапальна і антиоксидантна. Алкілування 5-R-1,2,4-триазол-3-тіолів проводили у безводному спиртовому середовищі або апротонних розчинниках. Встановлено, що заміна спиртового розчинника на апротонний збільшує кількісний вихід 5-R-1,2,4-триазол-3-тіоацетонітрилу; у апротонних розчинниках також практично не спостерігається присутність домішок продуктів лужного гідролізу. Синтезовано іміноефіри 2-(5-(2-, 3-, 4-метоксифеніл, 3,4,5-триметоксифеніл)-1Н-1,2,4-триазол-3-ілтіо)ацетатних кислот шляхом насичення 2-(5-(2-, 3-, 4-метоксифеніл, 3,4,5-триметоксифеніл)-1Н-1,2,4-триазол-3-ілтіо) ацетонітрилів струмом сухого хлористого водню у відповідному спиртовому середовищі при постійному охолодженні реакційної суміші до –5ºС. Методом ВЕРХ/ДМД-МС встановлено, що вихід цільового продукту залежить від витримки температурного режиму реакційної суміші. Структуру і індивідуальність молекул синтезованих речовин встановлено методами ^{1}Н ЯМР-спектроскопії і ВЕРХ/ДМД-МС.

СИНТЕЗ И ФИЗИКО-ХИМИЧЕСКИЕ СВОЙСТВА 2-(5-МЕТОКСИФЕНИЛ-1Н-1,2,4-ТРИАЗОЛ-3-ИЛТИО)АЦЕТОНИТРИЛОВ И ИХ ИМИНОЭФИРОВ

Ю.Г.Самелюк, А.Г.Каплаушенко

Ключевые слова: *1,2,4-триазолы; нитрилы; иминоэфиры*

С целью дальнейшего поиска биологически активных веществ среди 5-(2-, 3-, 4-метоксифенил, 3,4,5-триметоксифенил)-3-тио-1,2,4-триазолов и их производных были получены 10 новых соединений. Путем алкилирования 5-(2-, 3-, 4-метоксифенил, 3,4,5-триметоксифенил)-3-тио-1,2,4-триазолов галогеннитрилами были синтезированы ацетонитрилтио-1,2,4-триазолы. Первичный компьютерный фармакологический скрининг показал, что данный класс соединений может проявлять такие виды фармакологической активности, как противоопухолевая, противовоспалительная и антиоксидантная. Алкилирование 5-R-1,2,4-триазол-3-тиолов проводили в безводной спиртовой среде или апротонных растворителях. Установлено, что замена спиртового растворителя на апротонный увеличивает количественный выход 5-R-1,2,4-триазол-3-тиоацетонитрила; в апротонных растворителях также практически не наблюдается присутствие примесей продуктов щелочного гидролиза. Синтезированы иминоэфиры 2-(5-(2-, 3-, 4-метоксифенил, 3,4,5-триметоксифенил)-1Н-1,2,4-триазол-3-илтио)ацетатных кислот путем насыщения 2-(5-(2-, 3-, 4-метоксифенил, 3,4,5-триметоксифенил)-1Н-1,2,4-триазол-3-илтио)ацетонитрилов током сухого хлористого водорода в соответствующей спиртовой среде при постоянном охлаждении реакционной смеси до –5ºС. Методом ВЭЖХ/ДМД-МС установлено, что выход целевого продукта зависит от выдержки температурного режима реакционной смеси. Структура и индивидуальность молекул синтезированных веществ установлены методами ^{1}Н ЯМР-спектроскопии и ВЭЖХ/ДМД-МС.

A considerable interest is caused by the synthesis of biologically active compounds; among them there are substances several times exceeding their out-of-date analogues by action and, in addition, having lower indexes of acute and chronic toxicity. One of the classes of such compounds are derivatives of triazoles [1-6] used as original drugs with the antibacterial, neuroleptic, hypotensive and spasmolytic activity, and they are cardiac stimulants. The certain interest is also caused by synthetic medicines containing the nitrile group such as letrozole, anastrozole [6] used in modern medical practice as antitumor agents.

At the Physical and Colloidal Chemistry Department of the Zaporizhzhya State Medical University the investigations for searching the biologically active substances among 5-(2-, 3-, 4-methoxyphenyl and 3,4,5-trimethoxyphenyl)-3-thio-1,2,4-triazoles and their derivatives are being conducted. Earlier the corresponding 2-hydrazinocarbothioamides were obtained by interaction of hydrazides of 2-, 3-, and 4-methoxy- and 3,4,5-trimethoxybenzoic acids with potassium thiocyanate in the acidic medium; subsequently, 2-hydrazinocarbothioamides were cyclized when heating for 1 h under the action of 2 mole/l sodium hydroxide solution; 5-(2-, 3-, 4-methoxyphenyl)-1,2,4-triazole-3-thiols and 5-(3,4,5-trimethoxyphenyl)-1,2,4-triazole-3-thiol were obtained with the high yields [7] (Fig. 1).

At the present stage of our research acetonitrilothio-1,2,4-triazoles have been synthesized by alkylation of 5-(2-, 3-, 4-methoxyphenyl and 3,4,5-trimethoxyphenyl)-3-thio-1,2,4-triazoles with halogenonitriles; the primary computer pharmacological screening has shown that the class of compounds mentioned can show such types of the pharmacological activity as antitumor, anti-inflammatory and antioxidant ones.

Alkylation of 5-R-1,2,4-triazole-3-thiols was carried out in the medium of anhydrous alcohol or aprotic solvents with addition of Θ-chloroacetonitrile and the equimolar amount of alkali when heating the reaction mixture to the temperature of 60-65ºC; whereupon forming the precipitate of the expected reaction product was observed. After filtration the precipitate was washed with ether (Fig. 2).

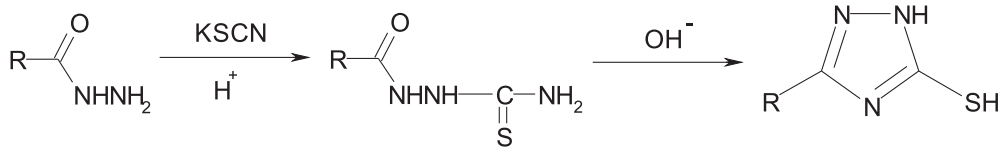
When studying the conditions of obtaining the target product the successful attempt of thiol alkylation without addition of an alkaline agent was made, and the corresponding hydrochloric salt was formed. The pure product was isolated with anhydrous sodium acetate. Thus, the samples of the compounds synthesized did not give the melting point depression.

Individuality of the corresponding substances has been proven by the method of HPLC/DAD-MS. Descriptions of ^{1}H NMR-spectra of the compounds obtained, some of their physical and chemical properties and the elemental composition are presented in Table 1 and 2.

It has been found that replacement of the alcoholic solvent by the aprotic one increases the quantitative yield of 5-R-1,2,4-triazol-3-thioacetonitrile; in aprotic solvents the presence of impurities of alkaline hydrolysis products is not practically observed (in the case of obtaining the target substances with the equimolar amount of alkali).

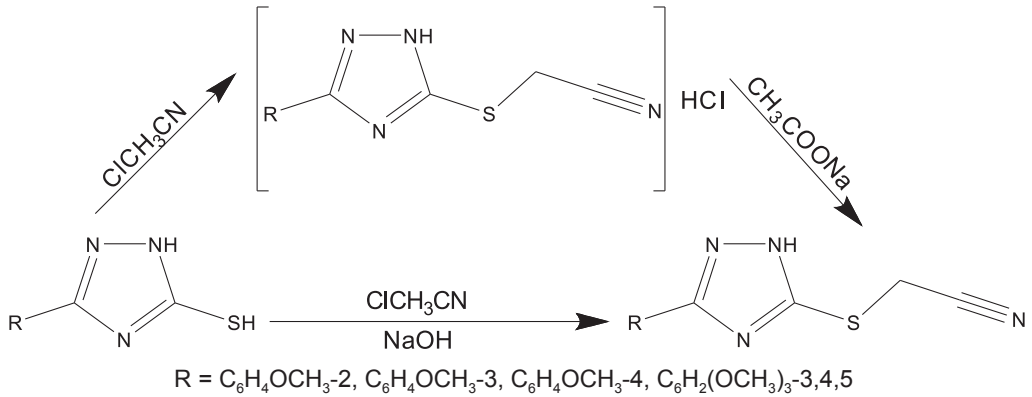
It should be noted that the highest yield was observed for compounds contained the 4-methoxyphenyl and 3,4,5-trimethoxyphenyl substituent at C^5 atom of the triazole ring (Table 1, compounds 3, 4). The lowest yield was observed for compound 2 (Table 1) with the 2-methoxyphenyl substituent.

In the ^{1}H NMR-spectra of the compounds of 2-(5-(2-,3-,4-methoxyphenyl, 3,4,5-trimethoxyphenyl)-1H-



R = $C_6H_4OCH_3$-2, $C_6H_4OCH_3$-3, $C_6H_4OCH_3$-4, $C_6H_2(OCH_3)_3$-3,4,5

Fig. 1. The scheme of obtaining 5-R-1,2,4-triazol-3-thiols.

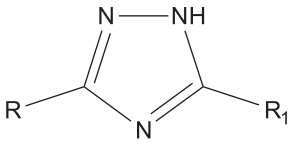


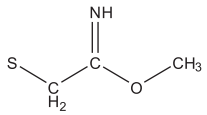
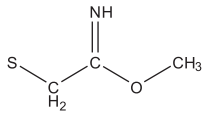
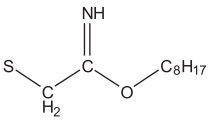
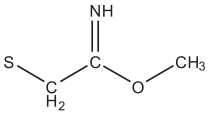
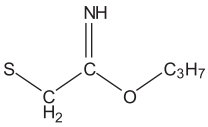
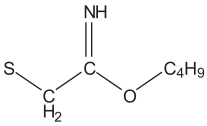
R = $C_6H_4OCH_3$-2, $C_6H_4OCH_3$-3, $C_6H_4OCH_3$-4, $C_6H_2(OCH_3)_3$-3,4,5

Fig. 2. The scheme of obtaining 5-R-1,2,4-triazol-3-thioacetonitriles.

Table 1

Physical and chemical constants of the compounds synthesized



No.	R_1	R_2^+	M.p., °C	Gross formula	Yield, %	^{1}H NMR (δ, m.p., TMS)
1	$C_6H_4OCH_3$-2	SCH_2CN	164-165	$C_{11}H_{10}N_4OS$	87	7.04-7.95 (4H, m, C_6H_4), 4.40 (2H, s, S-CH_2), 3.82 (3H, c, O-CH_3)
2	$C_6H_4OCH_3$-3	SCH_2CN	178-180	$C_{11}H_{10}N_4OS$	52	7.02-7.97 (4H, m, C_6H_4), 4.43 (2H, s, S-CH_2), 3.77 (3H, s, O-CH_3)
3	$C_6H_4OCH_3$-4	SCH_2CN	160-162	$C_{11}H_{10}N_4OS$	90	7.05-7.93 (4H, m, C_6H_4), 4.44 (2H, s, S-CH_2), 3.80 (3H, s, O-CH_3)
4	$C_6H_2(OCH_3)_3$-3,4,5	SCH_2CN	170-171	$C_{13}H_{14}N_4O_3S$	86	6.83 (2H, s, C_6H_2), 3.98 (2H, s, S-CH_2), 3.87 (3H, s, O-CH_3), 3.85 (3H, s, O-CH_3), 3.80 (3H, s, O-CH_3)
5	$C_6H_4OCH_3$-3		88-90	$C_{12}H_{14}N_4O_2S$	67	6.97-7.93 (4H, m, C_6H_4), 4.35 (2H, s, S-CH_2), 3.87 (3H, s, O-CH_3), 3.42 (3H, s, O-CH_3)
6	$C_6H_4OCH_3$-4		173-175	$C_{12}H_{14}N_4O_2S$	65	7,09-7.92 (4H, m, C_6H_4), 4.43 (2H, s, S-CH_2), 3.85 (3H, s, O-CH_3), 3.34 (3H, s, O-CH_3)
7	$C_6H_4OCH_3$-4		170-172	$C_{19}H_{28}N_4O_2S$	62	7,1-7.87 (4H, m, C_6H_4), 4.07 (2H, s, S-CH_2), 3.90 (3H, s, O-CH_3), 3.70 (2H, s, O-CH_2), 1.50 (12H, m, $(CH_2)_8$), 1.01 (3H, s, CH_3)
8	$C_6H_2(OCH_3)_3$-3,4,5		135-137	$C_{14}H_{18}N_4O_4S$	72	6.81 (2H, s, C_6H_4), 4.01 (2H, s, S-CH_2), 3.85 (3H, s, O-CH_3), 3.83 (3H, s, O-CH_3), 3.70 (3H, s, O-CH_3), 3.47 (3H, s, O-CH_3)
9	$C_6H_2(OCH_3)_3$-3,4,5		200-201	$C_{16}H_{22}N_4O_4S$	83	6.84 (2H, s, C_6H_2), 4.01 (2H, s, S-CH_2), 3.84 (3H, s, O-CH_3), 3.40 (3H, s, O-CH_3), 3.45 (3H, s, O-CH_3), 1.35 (2H, t, O-CH_2), 1.10 (2H, m, CH_2), 0.95 (3H, s, CH_3)
10	$C_6H_2(OCH_3)_3$-3,4,5		156-158	$C_{17}H_{24}N_4O_4S$	80	6.81 (2H, s, C_6H_2), 4.04 (2H, s, S-CH_2), 3.87 (3H, s, O-CH_3), 3.85 (3H, s, O-CH_3), 3.57 (3H, s, O-CH_3), 1.52 (2H, t, O-CH_2), 1.52 (4H, m, $(CH_3)_2$), 0.90 (3H, s, CH_3)

1,2,4-triazol-3-ylthio) acetonitriles synthesized and their acetimidates (iminoethers) the intense singlets are fixed in the range of 3.81-3.87 mp, which are the evidence of the presence of protons of the methoxy groups in the structure of all compounds [8].

One-proton singlets of the NH-group of N^2 atom of the 1H-1,2,4-triazole cycle at 13.48-13.55 mp are also recorded. Proton signals of the aromatic ring of the compounds with the 4-methoxyphenyl radical are interpreted by multiplet signals in the range of 7.92-7.98 mp, unlike singlet signals of the aromatic ring of the compounds with 3,4,5-trimethoxyphenyl substituents, which are fixed at 6.83-6.84 mp. The ^{1}H NMR-spectra are also characterized by the presence of the signal of protons of the methylene group in the range of 4.01-4.7 mp. The multiplet signals in the range of 0.90-3.46 mp confirm the presence of alkyl (alcohol) residues in iminoethers.

At the next stage of our research iminoethers (acetimidates) of 2-(5-(2-, 3-, 4-methoxyphenyl and 3,4,5-

Table 2

Elemental analysis of the compounds synthesized

Comp. No.	Found, %				Calculated, %			
	C	H	N	S	C	H	N	S
1	53.64	4.09	22.75	13.02	53.66	4.16	22.70	13.07
2	53.64	4.09	22.75	13.02	53.71	4.13	22.71	13.06
3	53.64	4.09	22.75	13.02	53.71	4.18	22.71	13.06
4	50.97	4.61	18.29	10.47	50.93	4.58	18.30	10.51
5	51.78	5.07	20.13	11.52	51.82	5.09	20.11	11.49
6	51.78	5.07	20.13	11.52	51.71	5.04	20.21	11.43
7	60.61	7.50	14.88	8.52	60.64	7.53	14.84	8.50
8	49.69	5.36	16.56	9.48	49.51	5.35	16.57	9.41
9	52.44	6.05	15.29	8.75	52.41	6.07	15.32	8.71
10	53.67	6.36	14.73	8.43	53.68	6.31	14.75	8.40

trimethoxyphenyl)-1H-1,2,4-triazole-3-ylthio)acetatic acids were synthesized by saturation of 2-(5- (2-, 3-, 4-methoxyphenyl and 3,4,5-trimethoxyphenyl)-1H-1,2,4-triazol-3-ylthio)acetonitriles with the flow of dry hydrogen chloride in the alcoholic medium when constant cooling the reaction mixture to -5ºC.

It has been found by the method of HPLC/DAD-MS that the qualitative yield of the target product depends on the maintenance of the temperature mode of the reaction mixture. When increasing the temperature the discrete intensive peak of the impurity of the unreacted initial 2-(5- (2-, 3-, 4-methoxyphenyl, 3,4,5-trimethoxyphenyl)-1H-1,2,4-triazol-3-ylthio) acetonitrilerises is observed on the chromatograms (Fig. 3).

The reaction proceeds in two stages [9] – at the first stage imine chlorides of acids are formed when affecting the corresponding 2-(5-R-1H-1,2,4-triazol-3-ylthio)acetonitrile with hydrogen chloride. Then at the second stage the substitution of the alcohol residue for chlorine with formation of hydrochloride of the corresponding iminoether takes place (Fig. 4).

Experimental Part

2-(5-(2-, 3-, 4-methoxyphenyl, 3,4,5-trimethoxyphenyl)-1H-1,2,4-triazol-3-ylthio)acetonitrile (1 – 4)

Method A. Boil the mixture of 0.01 Mole of 5-R-1,2,4-triazol-3-thiol and 0.02 Mole of chloroacetonitrile in 50 mL of propanol for 20 min (or until the complete dissolution of thiol); add anhydrous sodium acetate to the reaction mixture after complete cooling. Filter the precipitate of 2-(5-(2-, 3-, 4-methoxyphenyl, 3,4,5-trimethoxyphenyl)-1H-1,2,4-triazol-3-ylthio)acetonitrile, wash with diethyl ether and dry.

Method B. Boil the mixture of 0.01 Mole of 5-R-1,2,4-triazol-3-thiol and 0.02 Mol of sodium hydroxide in 50 ml of propanol for 20 min (or until the complete dissolution of thiol); add 0.02 Mole of chloro-

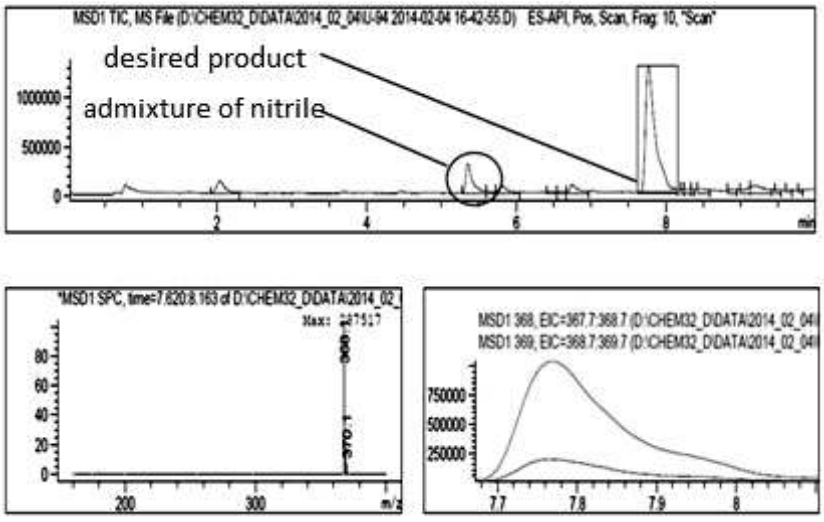
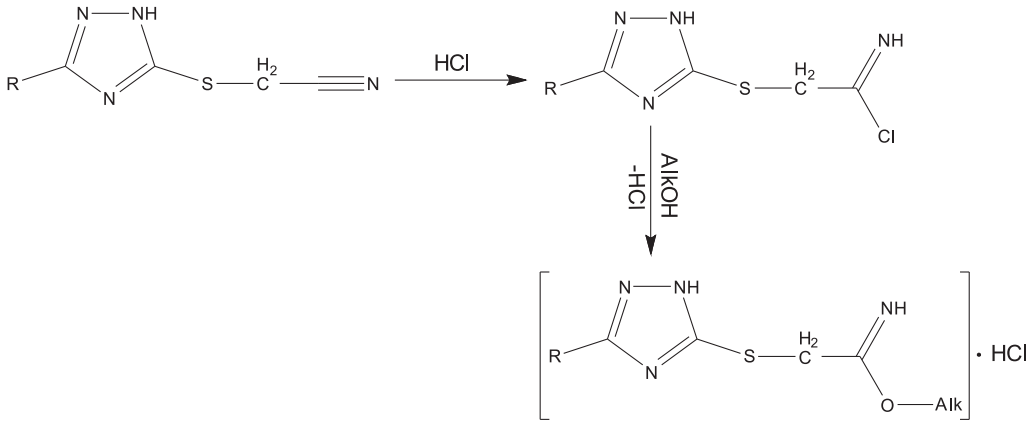


Fig. 3. The HPLC/DAD-MS-chromatogram of propyl-2-(5-(3,4,5-trimethoxyphenyl)-1H-1,2,4-triazole-3-ylthio)acetimidate obtained at the temperature of 2ºC.



R= $C_6H_4OCH_3$-2, $C_6H_4OCH_3$-3, $C_6H_4OCH_3$-4, $C_6H_2(OCH_3)_3$-3,4,5

Fig. 4. The scheme of obtaining 2-(5-R-1H-1,2,4-triazole-3-ylthio)acetimidates.

acetonitrile to the reaction mixture, boil the mixture for 20 min (until the neutral pH). Filter the primary precipitate of sodium chloride formed (from the solution heated). After complete cooling filter the precipitate of 2-(5 (2-, 3-, 4-methoxyphenyl, 3,4,5-trimethoxyphenyl)-1H-1,2,4-triazol-3-ylthio)acetonitrile formed, wash with diethyl ether and dry.

The yellowish crystalline substances obtained are soluble in alkaline solutions, as well as in organic solvents and solutions of mineral acids. For further analysis recrystallize these substances from ethanol.

Alkyl-2-(5-(2-, 3-, 4-methoxyphenyl, 3,4,5-trimethoxyphenyl)-1H-1,2,4-triazol-3-ylthio)acetimidate (5 – 9)

Place the solution of 0.01 Mole of 2-(5-(2-, 3-, 4-methoxyphenyl, 3,4,5-trimethoxyphenyl)-1H-1,2,4-triazol-3-ylthio)acetonitrile in 15 mL of chloroform and 25 mL of absolute alcohol into a Bunsen flask with the calcium chloride tube bound to its side tube. Close the flask with a stopper with the glass tube to the bottom of the flask connected to the drainage beakers. Cool the mixture in the desiccator filled with ice to the temperature of -5°C, and pass a flow of dry hydrogen chloride through the mixture, ultimately increment of hydrogen chloride is 2 mole with the excess of 1 mole related to the corresponding 2-(5-(2-, 3-, 4-methoxyphenyl, 3,4,5-trimethoxyphenyl)-1H-1,2,4-triazol-3-iltio)acetonitrile. After saturation with hydrogen chloride leave the reaction mixture in the refrigerator at the temperature of 0ºC. On the second day the light brown crystals, which are soluble in organic solvents and poorly soluble in water, are precipitated. Wash the substance with diethyl ether and dry. For further analysis recrystallize these substances from ethanol.

The study of some physical and chemical properties of the compounds synthesized (Table 1) was performed by the methods described in the State Pharmacopoeia of Ukraine (SPhU, 1st ed.).The melting point was determined by the capillary method (SPhU, 1st ed., 2.2.14) on a PTP(M) apparatus.

The molecular mass of substances and the presence of impurities were determined by the method of high-performance liquid chromatography under such conditions:

- device – Agilent 1260 Infinity HPLC System;
- software – OpenLAB;
- column – ∅4,6×30 mm, reversible phase Zorbax SB C18, 1,8 μm;
- column temperature – 40ºC;
- eluent A – H_2O – 0.1% HCOOH;
- eluent B – CH_3CN – 0.1% HCOOH;
- flow rate – 400 μL/min;
- gradient – linear from 5% to 100% of eluent B for 15 min;
- detector:

1) diode array (λ_1 = 210 nm; λ_2 = 254 nm);

2) Agilent 6120 single-quadrupole mass-spectrometer: ion source – API-ES; positive polarity; SIM mode; fragmentator – 10 V; drying gas – nitrogen (temperature – 300ºC, rate – 10 L/min); nebulizer pressure 40 psig; scanning in the range of m/z 160-1000.

The elemental composition of new compounds (Table 2) was determined using an ELEMENTAR vario EL cube elemental analyzer (sulfonamide as the standard).

^{1}H NMR spectra were recorded on a Varian VXR-300 spectrophotometer of nuclear magnetic resonance (DMSO-D_6 as a solvent, tetramethylsilane as the internal standard); the data were decoded with the ADVASP 143 software.

Conclusions

1. With the aim of further study of 1,2,4-triazoles with 2-, 3-, 4-methoxyphenyl, 3,4,5-trimethoxyphenyl substituent 10 new compounds have been synthesized.

2. For all compounds the preliminary biological screening has been carried out using the PASS ONLINE software.

3. The optimal synthetic conditions have been selected with the help of modern physical and chemical methods of analysis.

4. Individuality of the compounds synthesized has been determined, and their structure has been proven.

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ВЗАИМОДЕЙСТВИЕ ИОНОВ ПАЛЛАДИЯ(II) С АСПАРАГИНОМ В ВОДНЫХ РАСТВОРАХ

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Ключевые слова: комплексы палладия(II); аспарагин; константы образования комплексов; внутренняя координационная сфера комплексов

Методами электронной спектроскопии и pH-потенциометрии исследовано взаимодействие Pd(II) с аспарагином (Asn, HL) в водных растворах с физиологической концентрацией хлорид-ионов (0,15 моль/л KCl) в зависимости от соотношения Pd(II): Asn и pH. Установлено образование эквимолярных и бислигандных комплексов, в которых молекула Asn координирована к центральному иону металла бидентатно либо атомами азота аминогруппы и кислорода карбоксильной группы, либо атомами азота аминогруппы и депротонированной амидной группы. При бидентатной координации Asn в комплексах с соотношением металл : лиганд = 1 : 1 координационная сфера Pd(II) дополнена двумя хлорид-ионами. Состав внутренней координационной сферы комплексов Pd(II) охарактеризован на основании положения максимума полосы поглощения в электронных спектрах растворов. По программе PSEQUAD с учетом концентрации хлорид-ионов рассчитаны константы образования аспарагинатных комплексов Pd(II): lgβ $[PdLCl_2]^-$ = 19,36 (2); lgβ $[PdLH_{-1}Cl_2]^{2-}$ = 13,63 (1); lgβ $[PdL_2]$ = 25,99 (2); lgβ $[PdL_2H_{-1}]^-$ = 19,97 (4); lgβ $[PdL_2H_{-2}]^{2-}$ = 12,78 (4). На основании построенных диаграмм распределения равновесных концентраций комплексов показано, что при физиологических значениях pH и концентрации хлорид-ионов доминирующими в растворах являются комплексы $[PdLH_{-1}Cl_2]^{2-}$ при соотношении металл : лиганд = 1 : 1 и $[PdL_2H_{-1}]^-$, $[PdL_2H_{-2}]^{2-}$ при соотношении металл : лиганд = 1 : 2.

INTERACTION BETWEEN PALLADIUM(II) IONS AND ASPARAGINE IN AQUEOUS SOLUTIONS
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Key words: *complexes of Pd(II); asparagine; constants of complex formation; inner coordination sphere of complexes*
The interaction between Pd(II) and asparagines (Asn) in aqueous solutions with the physiological concentration of chloride ions (0.15 mol/L KCl) has been studied by election spectroscopy and pH potentiometry depending on the ratio of Pd(II):Asn and pH. The formation of equimolar and bisligand complexes has been found where the Asn molecule is coordinated to the central metal ion in a bidentate fashion either by the amine group nitrogen atom and the carboxyl group oxygen atom or by the nitrogen atoms of the amine group and the deprotonated amide group. In the case of bidentate coordination of Asn in complexes with the ratio of metal : ligand = 1 : 1, the Pd(II) coordination sphere is complemented by two chloride ions. The composition of the first coordination sphere of Pd(II) complexes has been characterized on the basis of the position of the absorption band maximum in the electronic spectra of solutions. By the PSEQUAD programme the formation constants of Pd(II) asparaginate complexes: lgβ $[PdLCl_2]^-$ = 19.36 (2); lgβ $[PdLH_{-1}Cl_2]^{2-}$ = 13.63 (1); lgβ $[PdL_2]$ = 25,99 (2); lgβ $[PdL_2H_{-1}]^-$ = 19.97 (4); lgβ $[PdL_2H_{-2}]^{2-}$ = 12.78 (4) have been calculated taking into account the concentration of chloride ions. Based on the equilibrium concentration distribution diagrams constructed for the complexes it has been shown that at the physiological values of pH and the chloride ion concentration the complexes $[PdLH_{-1}Cl_2]^{2-}$ in the ratio of metal : ligand = 1 : 1 and $[PdL_2H_{-1}]^-$, $[PdL_2H_{-2}]^{2-}$ in the ratio of metal : ligand = 1 : 2 predominate in solutions.

ВЗАЄМОДІЯ ІОНІВ ПАЛАДІЮ(II) З АСПАРАГІНОМ У ВОДНИХ РОЗЧИНАХ
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Ключові слова: *комплекси паладію(II); аспарагін; константи утворення комплексів; внутрішня координаційна сфера комплексів*
Методами електронної спектроскопії та pH-потенціометрії досліджено взаємодію Pd(II) з аспарагіном (Asn, HL) у водних розчинах з фізіологічною концентрацією хлорид-іонів (0,15 моль/л KCl) в залежності від співвідношення Pd(II):Asn та pH. Встановлено утворення еквімолярних та біслігандних комплексів, в яких молекула Asn може бути координована до центрального іону металу бідентатно або атомами азоту аміногрупи та кисню карбоксильної групи, або атомами азоту аміногрупи та депротонованої амідної групи. При бідентатній координації Asn в комплексах зі співвідношенням метал : ліганд = 1 : 1 координаційна сфера Pd(II) доповнена двома хлорид-іонами. Склад внутрішньої координаційної сфери комплексів Pd(II) охарактеризований на основі положення максимуму смуги поглинання в електронних спектрах розчинів. За програмою PSEQUAD із врахуванням концентрації хлорид-іонів розраховано константи утворення аспарагінатних комплексів Pd(II):): lgβ $[PdLCl_2]^-$ = 19,36 (2); lgβ $[PdLH_{-1}Cl_2]^{2-}$ = 13,63 (1); lgβ $[PdL_2]$ = 25,99 (2); lgβ $[PdL_2H_{-1}]^-$ = 19,97 (4); lgβ $[PdL_2H_{-2}]^{2-}$ = 12,78 (4). На підставі побудованих діаграм розподілу рівноважних концентрацій комплексів показано, що за фізіологічних значень pH та концентрації хлорид-іонів домінуючими в розчинах є комплекси $[PdLH_{-1}Cl_2]^{2-}$ при співвідношенні метал : ліганд = 1 : 1 та $[PdL_2H_{-1}]^-$, $[PdL_2H_{-2}]^{2-}$ при співвідношенні метал : ліганд = 1 : 2.

Широкий интерес к комплексным соединениям металлов платиновой группы с биологически активными лигандами вызван необходимостью создания новых более эффективных и менее токсичных противоопухолевых препаратов, чем применяемые в онкологической практике соединения платины: цисплатин, карбоплатин и оксалиплатин [1-3]. Выявленная для соединений Pd(II) значительно меньшая токсичность по сравнению с комплексами Pt(II) указывает на перспективность их применения в медицине [4-6]. Изучение комплексов Pd(II), плоско-квадратный координационный узел которых образован различными донорными атомами функциональных групп биологических молекул, позволит расширить представления о биокоординационной химии металлов платиновой группы.

Внимание к аминокислотам в качестве лигандов обусловлено как их биологической ролью, так и разнообразием типов соединений с этими полифункциональными лигандами. Наличие в составе ряда аминокислот кроме амино- и карбоксильной групп дополнительных функциональных групп обусловливает возможность образования комплексов с различным способом координации лиганда к иону металла. Одной из таких аминокислот является аспарагин (Asn), содержащий боковую амидную группу, которая может координироваться к Pd(II) при депротонировании амидного атома азота.

В литературе приведены данные о синтезе комплекса Pd(II) с Asn, в котором две молекулы лиганда координированы к центральному иону металла бидентатно донорными атомами азота аминогруппы и кислорода карбоксильной группы [7]. В работах, посвященных исследованию комплексообразования Pd(II) с Asn в растворах, выявлено образование комплексов при координации лиганда атомами азота аминогруппы и депротонированной амидной группы [8-10]. Из анализа литературных данных по исследованию устойчивости аспарагинатных комплексов Pd(II) (таблица) следует, что для расчета констант образования комплексов применяли, в основном, данные pH-потенциометрического титрования, обработка которых предполагала априорный выбор состава образующихся комплексов. Более информативным для определения состава и строения комплексов Pd(II) является метод электронной спектроскопии [11, 12].

Таким образом, целью данной работы являлось исследование комплексообразования Pd(II) с Asn методами спектрофотометрии и pH-потенциометрии в растворах с физиологической концентрацией хлорид-ионов (0,15 моль/л KCl), включающее определение состава внутренней координационной сферы комплексов и расчет констант образования комплексов с одновременной обработкой данных двух методов.

Сложность изучения комплексообразования Pd(II) связана с медленным гидролизом $[Pd(H_2O)_4]^{2+}$ в растворах при pH>1 с образованием малорастворимых гидроксокомплексов [13]. При использовании в качестве исходных соединений $PdCl_2$ или K_2PdCl_4 в растворе образуются хлороаквакомплексы состава $[PdCl_{4-n}(H_2O)_n]^{(n-2)-}$, где n = 0-3 [14]. Соотношение равновесных концентраций различных по составу хлороаквакомплексов в растворе зависит от концентрации хлорид-ионов, а образующиеся комплексы Pd(II) могут содержать в координационной сфере помимо донорных атомов лиганда молекулы воды или хлорид-ионы.

Таблица

Константы образования комплексов Pd(II) с Asn

Исходное соединение Pd(II)	Метод*	Среда, моль/л	T, (°C)	Состав комплекса	lgβ	Источник
Na_2PdCl_4	а	~0,02	27	ML_2	15,12	[8]
$Pd(ClO_4)_2$	а, б	3,0 $NaClO_4$	25	MLH MLH_{-1} MLHCl $MLH_{-1}Cl$	12,11 9,1 18,29 17,0	[9]
$PdCl_2$	а	0,1 $NaClO_4$	25	ML ML_2	9,15 17,65	[10]
K_2PdCl_4	а, с	0,15 KCl	20	HL $[H_2L]^+$ $[PdLCl_2]^-$ $[PdLH_{-1}Cl_2]^{2-}$ $[PdL_2]$ $[PdL_2H_{-1}]^-$ $[PdL_2H_{-2}]^{2-}$	8,89 (3)** 11,30 (3) 19,36 (2) 13,63 (1) 25,99 (2) 19,97 (4) 12,78 (4)	Наши данные

Методы: а – pH-потенциометрия, б – Cl^- – селективный электрод, с – спектрофотометрия.
** – в скобках приведены стандартные отклонения последней значущей цифры величины lgβ.

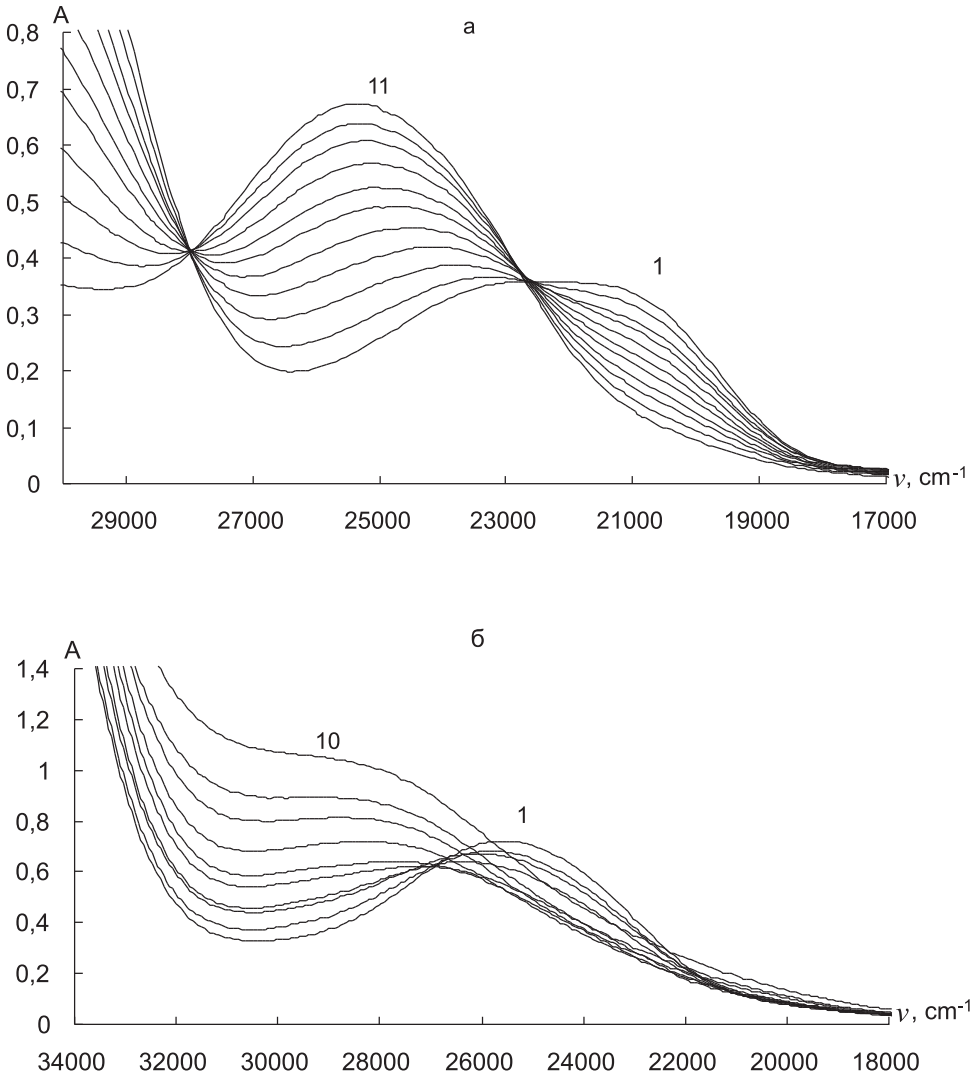


Рис. 1. ЭСП-системы K_2PdCl_4-Asn в 0,15 моль/л KCl: **а)** ($C_{Pd(II)}$ = 2,5 · 10^{-3}моль/л, C_{Asn} = 4 · 10^{-4} – 2,6 · 10^{-3} моль/л); pH: 1 – 3,58; 2 – 3,37; 3 – 3,22; 4 – 3,13; 5 – 3,05; 6 – 2,98; 7 – 2,93; 8 – 2,89; 9 – 2,84; 10 – 2,81; 11 – 2,77; **б)** ($C_{Pd(II)}$ = C_{Asn}=2,5 · 10^{-3} моль/л); pH: 1 – 4,82; 2 – 5,31; 3 – 5,52; 4 – 5,67; 5 – 5,96; 6 – 6,09; 7 – 6,31; 8 – 6,44; 9- 6,55; 10 – 6,61.

Поэтому расчет констант образования комплексов Pd(II) необходимо проводить с учетом констант образования хлороаквакомплексов, концентрации хлорид-ионов в растворе и наличия хлорид-ионов в составе образующихся комплексов. Ранее с включением в стартовую модель хлороаквакомплексов Pd(II) были рассчитаны константы образования комплексов Pd(II) с глицином, L-аланином и L-гистидином [15].

В данной работе проведен расчет констант образования комплексов Pd(II) с Asn по формуле (1):

$$\beta = [M_xL_yH_zCl_q]/[M]^x[L]^y[H]^z[Cl]^q \qquad (1)$$

В расчетах использовали константы образования хлороаквакомплексов палладия(II): $lg\beta PdCl^+$ = 4,47; $lg\beta PdCl_2$ = 7,76; $lg\beta PdCl_3^-$ = 10,17; и $lg\beta PdCl_4^{2-}$ = 11,54 (I = 1 моль/л $HClO_4$ + $NaClO_4$, T = 25°C) [14].

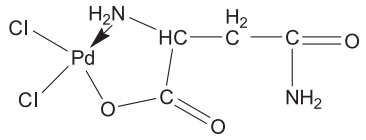
Определение состава внутренней координационной сферы комплексов Pd(II) с Asn проводили по сопоставлению положения максимума d-d полосы в электронных спектрах поглощения (ЭСП) ($\nu_{эксп.}$) со значением ($\nu_{рассч.}$), рассчитанным по формуле (2):

$$\nu_{рассч.}=\Sigma n_i N_I \qquad (2),$$

где n_i – количество донорных атомов каждого типа, ν_i – величина инкремента для донорных атомов каждого типа: ν(Cl) = 5170 $см^{-1}$, ν($N_{амин.}$) = 8460 $см^{-1}$, ν($O_{карбокс.}$) = 6770 $см^{-1}$, ν($N_{амид.}$) = 9090 $см^{-1}$ [12].

В ЭСП серии растворов K_2PdCl_4 (C = 0,0025 моль/л) в 0,15 моль/л KCl при увеличении концентрации Asn (0,0004 – 0,0026 моль/л) наблюдался сдвиг максимума полосы поглощения с $\nu_{эксп.}$ = 21400 $см^{-1}$, соответствующей исходным хлороаквакомплексам, к 25300 $см^{-1}$, что может свидетельствовать об образовании комплекса с составом хромофора [Pd $N_{амин.}$ $O_{карбокс.}$ 2Cl], для которого $\nu_{рассч.}$ = 25600 $см^{-1}$ (рис. 1а). Таким образом, состав внутренней координационной сферы образовавшегося комплекса может быть сформирован атомами азота амино-

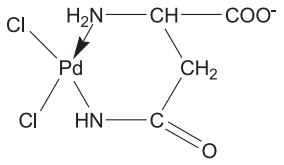
группы и кислорода карбоксильной группы Asn и двумя хлорид-ионами. При расчете константы образования комплекса состава $[PdLCl_2]^-$ использовали значения величин оптических плотностей на 22 частотах на участках 26000-24500 $см^{-1}$ и 21300-20500 $см^{-1}$ и значения pH серии растворов (таблица).



$[PdLCl_2]^-$

При увеличении pH растворов с соотношением K_2PdCl_4:Asn = 1:1 от 4,82 до 6,60 наблюдалось дальнейшее смещение максимума полосы поглощения от 25300 $см^{-1}$ до 28200 $см^{-1}$ (рис. 1б). Такие изменеия в ЭСП могут свидетельствовать об образовании комплекса с составом хромофора [Pd $N_{амин.}$ $N_{амид.}$ 2Cl] со значением $\nu_{рассч.}$ = 27900 $см^{-1}$ при координации молекулы Asn атомами азота аминогруппы и депротонированной амидной группы.

По значениям оптических плотностей на 21 частоте в области 30400-24600 $см^{-1}$ и pH серии растворов рассчитана величина константы образования комплекса состава $[PdLH_{-1}Cl_2]^{2-}$ (таблица).

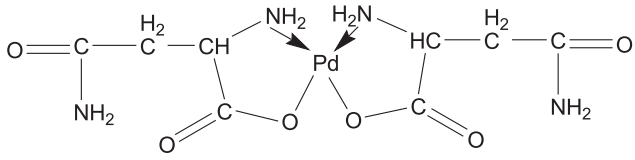


$[PdLH_{-1}Cl_2]^{2-}$

На основании значений констант образования комплексов эквимолярного состава рассчитана константа равновесия реакции депротонирования амидной группы лиганда:

$[PdLCl_2]^- = [PdLH_{-1}Cl_2]^{2-} + [H]^+$ ($pK_{H(1)}$ = 5,73).

В ЭСП растворов системы K_2PdCl_4:Asn = 1:2 при увеличении pH от 2,89 до 7,46 можно выделить три области pH, для которых наблюдалось последовательное смещение максимума полосы поглощения, свидетельствующее об образовании трех комплексов с различным составом хромофора (рис. 2). В области pH 3,28-5,27 смещение максимума полосы поглощения от 25300 $см^{-1}$ к 31000 $см^{-1}$ может быть обусловлено образованием бислигандного комплекса с составом хромофора [Pd $2N_{амин.}$ $2O_{карбокс.}$] со значением $\nu_{рассч.}$ = 30500 $см^{-1}$ (рис. 2а). Комплекс с таким составом хромофора образуется при бидентатной координации к Pd(II) двух молекул Asn атомами азота аминогруппы и кислорода карбоксильной группы. По величинам оптических плотностей серии растворов на 20 частотах в интервалах 32400-30200 $см^{-1}$ и 26600-24600 $см^{-1}$ и значениям pH рассчитана константа образования комплекса состава $[PdL_2]$ (таблица).

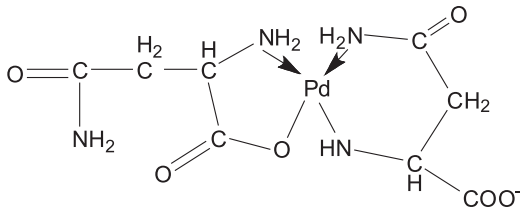


$[PdL_2]$

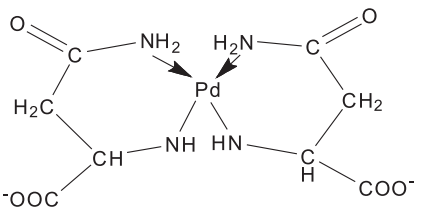
В дальнейшем строение комплексов приводится без учета возможности образования транс-изомеров.

Дальнейшее смещение максимума полосы поглощения к 32600 $см^{-1}$ при повышении pH от 5,58 до 6,48 и к 34000 $см^{-1}$ при повышении pH от 6,99 до 8,21 может свидетельствовать о последовательной координации депротонированных амидных групп двух молекул лиганда (рис. 2б и 2в). Такое предположение сделано на основании сопоставления $\nu_{эксп.}$ с $\nu_{рассч.}$, равным 32800 $см^{-1}$ для хромофора [Pd $2N_{амин.}$ $O_{карбокс.}$ $N_{амид.}$] и 35100 $см^{-1}$ для хромофора [Pd $2N_{амин.}$ $2N_{амид.}$]. Отличие значения $\nu_{рассч.}$ от $\nu_{эксп.}$ для хромофора [Pd $2N_{амин.}$ $2N_{амид.}$] может быть связано как со сложностью точного определения положения максимума очень широкой полосы поглощения, так и, возможно, недостаточно точным значением $\nu(N_{амид.})$.

При расчете константы образования комплекса $[PdL_2H_{-1}]^-$ использовали оптические плотности на 15 частотах в интервале 33200-30900 $см^{-1}$ и на 21 частоте в интервале 36000-31600 для комплекса $[PdL_2H_{-2}]^{2-}$ (таблица).



$[PdL_2H_{-1}]^-$



$[PdL_2H_{-2}]^{2-}$

Рассчитаны константы равновесия реакций последовательного депротонирования амидных групп двух координированных молекул Asn:

$[PdL_2] = [PdL_2H_{-1}]^- + [H]^+$ ($pK_{H(2)}$ = 6,02);

$[PdL_2H_{-1}]^- = [PdL_2H_{-2}]^{2-} + [H]^+$ ($pK_{H(3)}$ = 7,19).

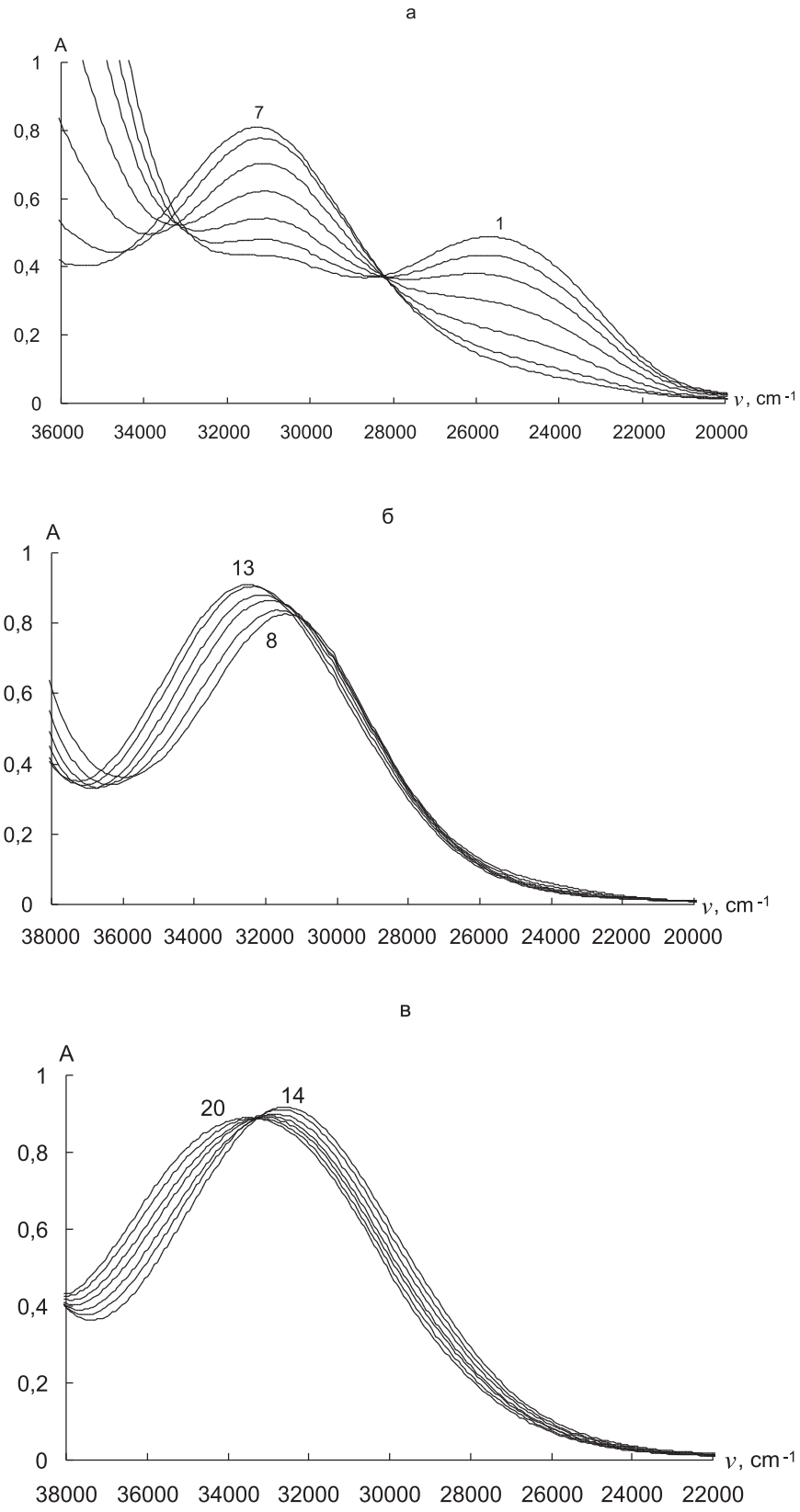


Рис. 2. ЭСП-системы K_2PdCl_4:Asn = 1:2 в 0,15 моль/л KCl ($C_{Pd(II)}$ = 2,5 · 10^{-3}, C_{Asn} = 5 · 10^{-3}моль/л; pH: **а)** 1 – 3,28; 2 – 3,44; 3 – 3,66; 4 – 3,99; 5 – 4,40; 6 – 4,91; 7 – 5,27; **б)** 8 – 5,58; 9 – 5,82; 10 – 6,12; 11 – 6,31; 12 – 6,59; 13 – 6,84; **в)** 14 – 6,99; 15 – 7,17; 16 – 7,37; 17 – 7,56; 18 – 7,71; 19 – 7,95; 20 – 8,21.

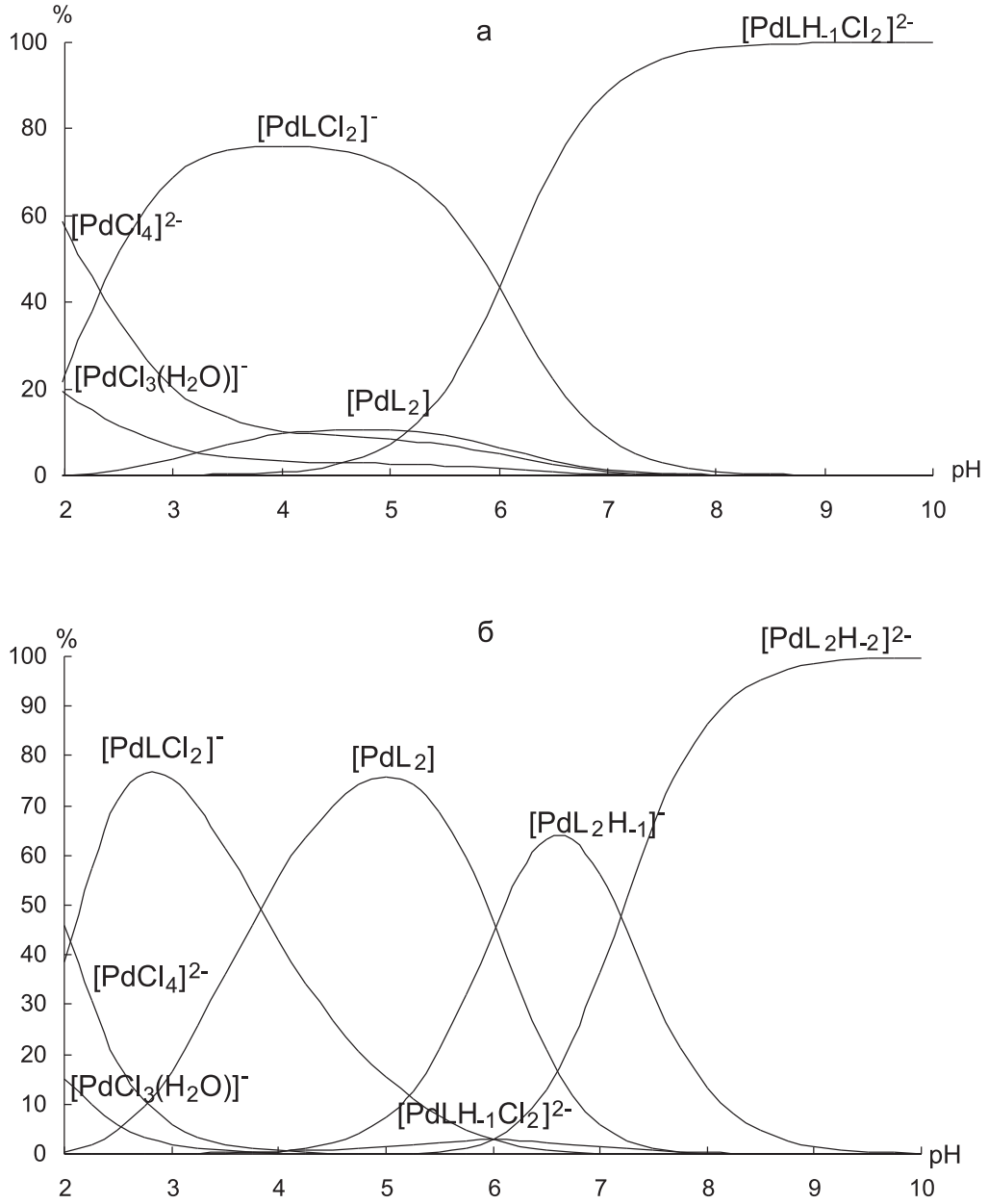


Рис. 3. Диаграммы распределения равновесных концентраций комплексов Pd(II) с Asn в 0,15 моль/л KCl:
а) K_2PdCl_4 : Asn = 1:1 ($C_{Pd(II)} = C_{Asn}$=2,5 · 10^{-3} моль/л); **б)** K_2PdCl_4 : Asn = 1:2 ($C_{Pd(II)}$ = 2,5 · 10^{-3} моль/л, C_{Asn} = 5 · 10^{-3} моль/л).

Из диаграмм распределения равновесных концентраций комплексов следует, что при физиологических значениях pH (~7.4) и концентрации хлорид-ионов (0,15 моль/л) доминирующими комплексами являются $[PdLH_{-1}Cl_2]^{2-}$ в системе K_2PdCl_4:Asn = 1:1 и $[PdL_2H_{-1}]^-$, $[PdL_2H_{-2}]^{2-}$ в системе K_2PdCl_4:Asn = 1:2 (рис. 3).

Экспериментальная часть

В качестве исходных веществ использовали K_2PdCl_4, полученный по методике [10], и L-аспарагин (Asn, HL) марки «ч. д. а».

pH-Потенциометрическое титрование Asn проводили 0,1 моль/л раствором КОН, свободным от карбонатов, при 20±0,1°С. Концентрация Asn в исходном растворе составляла 0,005 моль/л, начальный объем титруемого раствора – 25 мл.

Изучение комплексообразования в системе K_2PdCl_4 – Asn проводили методами pH-потенциометрии и спектрофотометрии для серии растворов с постоянной концентрацией K_2PdCl_4 (0,0025 моль/л) и переменной концентрацией лиганда (0,0004-0,0026 моль/л). Исследовали также серии растворов, содержащих K_2PdCl_4 и Asn в соотношении 1:1 и 1:2 в зависимости от количества добавленного КОН. Все исследуемые растворы имели постоянную ионную силу (I = 0,15 моль/л KCl), что соответствовало физиологической концентрации хлорид-ионов. Измерения pH и запись электронных спектров поглощения (ЭСП) серии растворов, содержащих K_2PdCl_4 и Asn, проводили через 24 часа после приготовления.

pH Растворов измеряли на pH-метре 827 pH lab фирмы «Metrohm» при 20±1^{0}С. Калибровку pH-мет-

ра проводили с использованием стандартных буферных растворов с pH 1,68; 4,01; 6,86 и 9,18 с учетом зависимости их pH от температуры. Электронные спектры поглощения записывали на спектрофотометре Specord-M40 в кварцевой кювете (l=1 см). Константы образования протонированых форм Asn, константы образования комплексов Pd(II) с Asn и их равновесные концентрации рассчитывали по программе PSEQUAD [16] с использованием данных спектрофотометрии и pH-потенциометрии.

Выводы

1. В результате проведенного методами спектрофотометрии и pH-потенциометрии исследования комплексообразования K_2PdCl_4 с Asn в растворах с физиологической концентрацией хлорид-ионов (0,15 моль/л KCl) установлено образование комплексов эквимолярного и бислигандного составов. Показано, что координация молекул Asn к центральному иону металла в комплексах может осуществляться как атомами азота аминогруппы и кислорода карбоксильной группы, так и атомами азота аминогруппы и депротонированной амидной группы.

2. Рассчитаны константы образования комплексов Pd(II) с Asn с учетом концентрации хлорид-ионов в растворе и наличия хлорид-ионов в составе образующихся комплексов.

3. На основании построенных диаграмм распределения равновесных концентраций комплексов установлен состав доминирующих комплексов Pd(II) с Asn, образующихся в растворах при близких к физиологическим значениям pH и концентрации хлорид-ионов.

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ЗРУЧНИЙ ПІДХІД ДО СИНТЕЗУ ЗАМІЩЕНИХ 2-(МЕТИЛАМІНО)ХІНОЛІН-3-КАРБОКСАМІДІВ

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Ключові слова: конденсація; піримідо[4,5-b]хінолін-2,4(1H,3H)-діони; 5-деазаалоксазини; 2-(метиламіно)хінолін-3-карбоксаміди

Робота присвячена синтезу нових похідних 2-(метиламіно)хінолін-3-карбоксамідів шляхом гідролітичного розщеплення піримідинового циклу в піримідо[4,5-b]хінолін-2,4(1H,3H)-діонах. В основі даного підходу лежить взаємодія доступного 1,3-диметил-6-піперидиніл-5-формілурацилу з ароматичними амінами, в результаті чого було синтезовано ряд заміщених похідних піримідо[4,5-b]хінолін-2,4(1H,3H)-діонів з високими виходами. Реакція 1,3-диметил-6-піперидиніл-5-формілурацилу з орто- та пара-заміщеними ароматичними амінами перебігає регіоселективно з утворенням похідних 1,3-диметилпіримідо[4,5-b] хінолін-2,4(1H,3H)-діонів з замісниками в положеннях 7 та 9 гетероциклу. Використання в реакції мета-метокси- чи мета-фтороаніліну приводить до суміші 6- та 8-заміщених 1,3-диметилпіримідо[4,5-b] хінолін-2,4(1H,3H)-діонів, які при дії лугу розщеплюються до відповідних амідів хінолін-3-карбонової кислоти. Отриману суміш 2-(метиламіно)хінолін-3-карбоксамідів вдалося розділити за допомогою хроматографії на колонці. Застосування методики кореляційної спектроскопії ЯМР (NOESY, COSY, HSQC, HMBC) дозволило достовірно встановити будову продуктів реакції конденсаціїї похідних 1,3-диметил-5-формілурацилу з ароматичними амінами. При нагріванні до ~120°C можлива аміно-імінна прототропія, яка полегшує син-анти-таутомерію в положенні 6 піримідинового кільця, що приводить до циклізації в 1,3-диметилпіримідо[4,5-b]хінолін-2,4(1H,3H)-діон. Структура отриманих сполук чітко доведена з використанням методів ЯМР ^{1}H, ^{13}C-спектроскопії, хроматомас-спектрометрії та елементного аналізу.

A CONVENIENT APPROACH TO THE SYNTHESIS OF SUBSTITUTED 2-(METHYLAMINO)QUINOLINE-3-CARBOXAMIDES

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Key words: *condensation; pyrimido[4,5-b]quinoline-2,4(1H,3H)-dione; 5-deazaalloxazine; 2-(methylamino)quinoline-3-carboxamide*

The paper is devoted to the synthesis of new 2-(methylamino)quinoline-3-carboxamide derivatives by the pyrimidine cycle hydrolytic cleavage of pyrimido[4,5-b]quinoline-2,4(1H,3H)-diones. These later were synthetized in high yields by interaction of available 1,3-dimethyl-6-piperidinyl-5-formyluracil with aromatic amines. The reactions with participation of ortho- and para-substituted aromatic amines proceed regioselectively to give 1,3-dimethylpyrimido[4,5-b] quinoline-2,4(1H,3H)-diones with substituents in positions 7 and 9 of the heterocycle. The use of metha-methoxy- and metha-fluoroaniline leads to the mixture of 6- and 8-substituted 1,3-dimethylpyrimido[4,5-b]quinoline-2,4(1H,3H)-diones, which under basic conditions were cleaved into amides of quinoline-3-carboxylic acid. The mixture of 2-(methylamino)quinoline-3-carboxamides obtained was separated by column chromatography. Application of the correlation NMR spectroscopy method (NOESY, COSY, HSQC, HMBC) has allowed to reliably determine the structures of 1,3-dimethyl-5-formyluracil derivatives with aromatic amines condensation products. The amino-imino prototropic tautomerism occurs at 120°C. It facilitates syn-anti-isomerization promoted cyclization and leads to 1,3-dimethylpyrimido[4,5-b]quinoline-2,4(1H,3H)-dione. The structure of the compounds obtained has been proven using the methods of NMR ^{1}H, ^{13}C-spectroscopy, mass-spectra and elemental analysis.

УДОБНЫЙ ПОДХОД К СИНТЕЗУ ЗАМЕЩЕННЫХ 2-(МЕТИЛАМИНО)-ХИНОЛИН-3-КАРБОКСАМИДОВ

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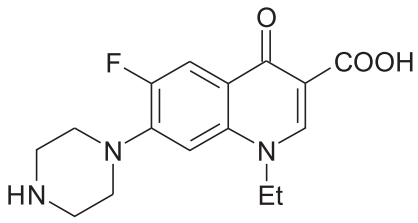
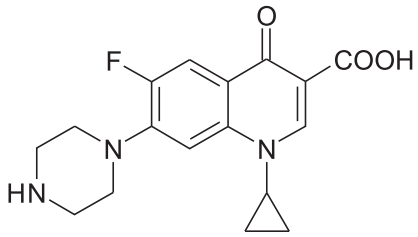
Ключевые слова: *конденсация; пиримидо[4,5-b]хинолин-2,4(1H,3H)-дионы; 5-деазааллоксазины; 2-(метиламино)хинолин-3-карбоксамиды*

Работа посвящена синтезу новых производных 2-(метиламино)хинолин-3-карбоксамидов путем гидролитического расщепления пиримидинового цикла в пиримидо[4,5-b]хинолин-2,4(1H,3H)-дионах. В основе данного подхода лежит взаимодействие доступного 1,3-диметил-6-пиперидинил-5-формилурацила с ароматическими аминами, в результате чего было синтезировано ряд замещенных производных пиримидо[4,5-b] хинолин-2,4(1H,3H)-дионов с высокими выходами. Реакция 1,3-диметил-6-пиперидинил-5-формилурацила с орто- и пара-замещенными ароматическими аминами протекает региоселективно с образованием производных 1,3-диметилпиримидо[4,5-b]хинолин-2,4(1H,3H)-дионов с заместителями в положениях 7 и 9 гетероцикла. Использование в реакции мета-метокси- или мета-фторанилина приводит к смеси 6- и 8-замещенных 1,3-диметилпиримидо[4,5-b]хинолин-2,4(1H,3H)-дионов, которые при действии щелочи расщепляются до соответствующих амидов хинолин-3-карбоновой кислоты. Полученную смесь 2-(метиламино)хинолин-3-карбоксамидов удалось разделить с помощью хроматографии на колонке. Использование методики корреляционной спектроскопии ЯМР (NOESY, COSY, HSQC, HMBC) позволило однозначно установить строение продуктов реакции конденсации производных 1,3-диметил-5-формилурацила с ароматическими аминами. При нагревании до ~120°C возможна амино-иминная прототропия, которая облегчает син-анти-таутомерию в положении 6 пиримидинового кольца, что приводит к циклизации в 1,3-диметилпиримидо[4,5-b]хинолин-2,4(1H,3H)-дион. Структура полученных соединений строго доказана с использованием методов ЯМР ^{1}H, ^{13}C-спектроскопии, хроматомасс-спектрометрии и элементного анализа.

Інтенсивний пошук антимікробних препаратів серед похідних 4-хінолін-3-карбонових кислот у 80-х роках минулого століття привів до отримання високоефективних антибіотиків – норфлоксацину та ципрофлоксацину.



Норфлоксацин



Ципрофлоксацин

Ці препарати володіють широким спектром антимікробної дії, а також низькою здатністю індукувати у бактерій резистентність до антибіотиків. Особливо важливі ці речовини при лікуванні інфекцій, викликаних штамами, стійкими до інших лікарських засобів.

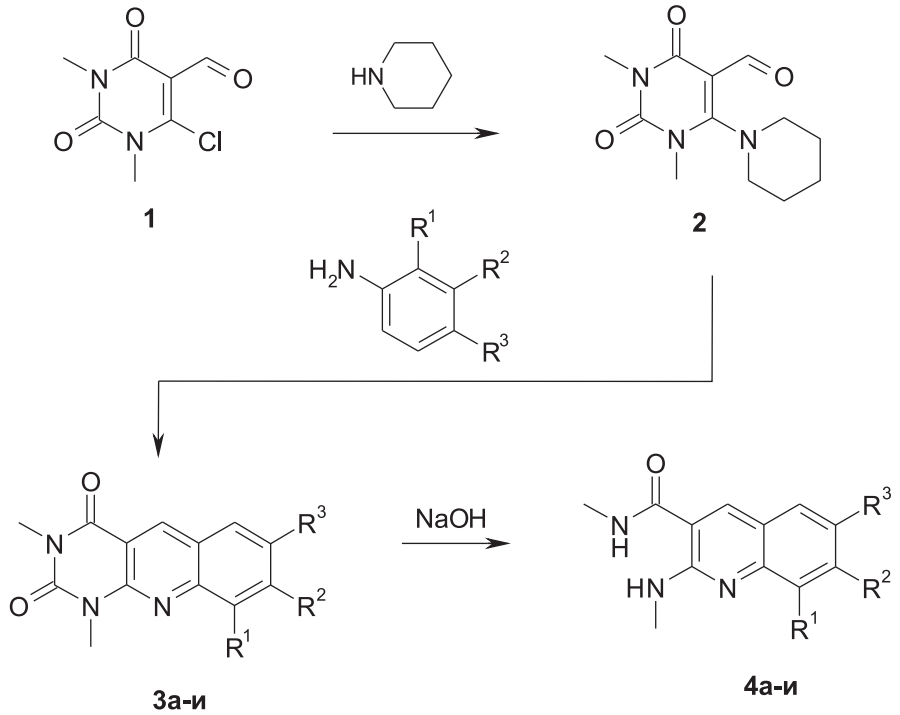
Інтерес до похідних хінолін-3-карбонових кислот не зменшується і до сьогоднішнього дня. В ряду N-алкілзаміщених 4-оксохінолін-3-карбонових кислот виявлені сполуки, що проявляють антимікробну активність [1-6], антимікобактеріальну дію [7-9], а також є інгібіторами інтегрази ВІЛ [10] та вірусу гепатиту С [11].

В останні роки увагу дослідників почали привертати заміщені 2-амінохінолін-3-карбоксаміди як модулятори калієвих каналів KCNQ 2/3 [12], які можуть бути перспективними для лікування неврологічних захворювань. Крім цього, ці сполуки є потенційними інгібіторами β-секретази (BACE) [13], ацетилхолінестерази [14], тирозинкінази [15], а також агоністами каннабіноїдних рецепторів CB1 та CB2 [16-19].

Одним із поширених способів синтезу 2-амінозаміщених хінолін-3-карбоксамідів є конденсація *о*-амінокарбонільних сполук з похідними ціаноцтової кислоти (модифікація синтезу Фрідлендера) [20-22].

Нами запропонований зручний спосіб синтезу 2-(метиламіно)хінолін-3-карбоксамідів **4а-и** (схема 1), з використанням піримідо[4,5-*b*]хінолін-2,4(1*H*,3*H*)-діонів (5-деазаалоксазинів) **3а-и**.

Слід відмітити, що в літературі описані методи синтезу сполук **3** з використанням 6-ариламіно-



3, 4: R^1 = OMe, R^2 = R^3 = H (а); R^1 = F, R^2 = R^3 = H (б); R^1 = R^3 = H, R^2 = CF_3 (в); R^1 = H, R^2 = R^3 = OMe (г); R^1 = R^3 = OMe, R^2 = H (д); R^1 = R^2 = H, R^3 = Me (е); R^1 = R^2 = H, R^3 = Et (ж); R^1 = R^2 = H, R^3 = *n*-Bu (з); R^1 = R^2 = H, R^3 = OEt (и).

Схема 1

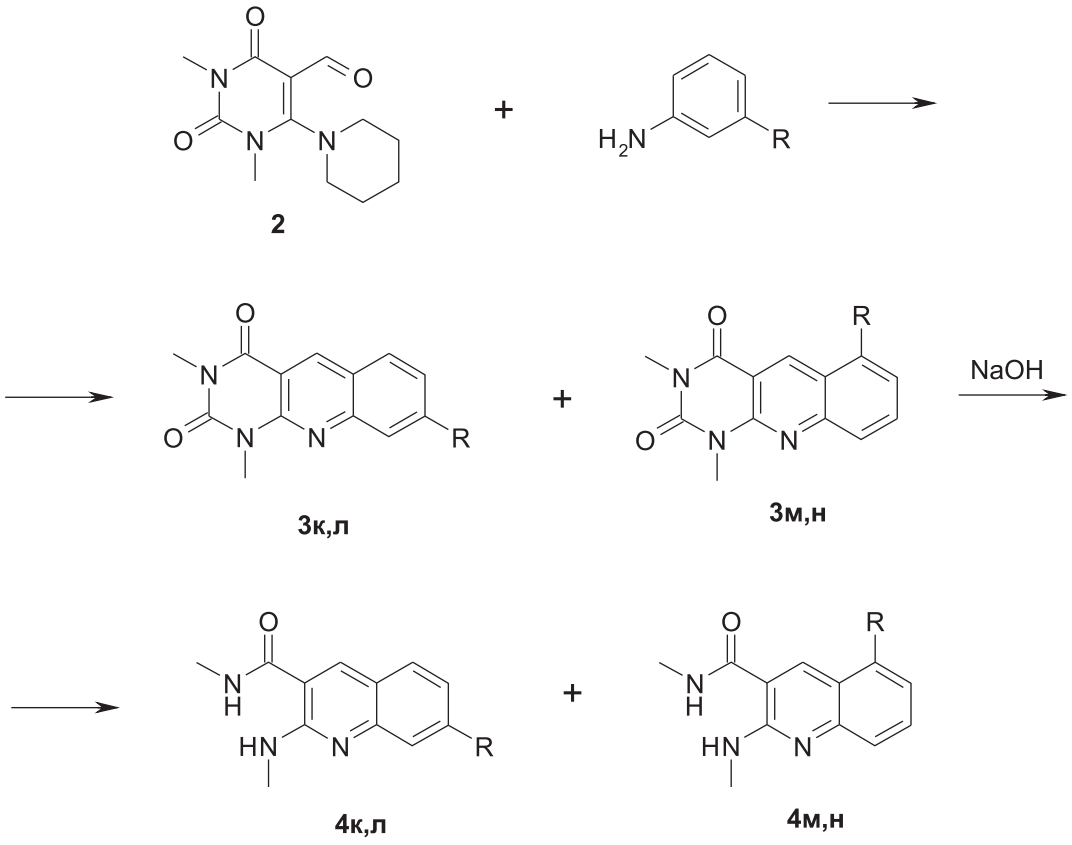


Схема 2

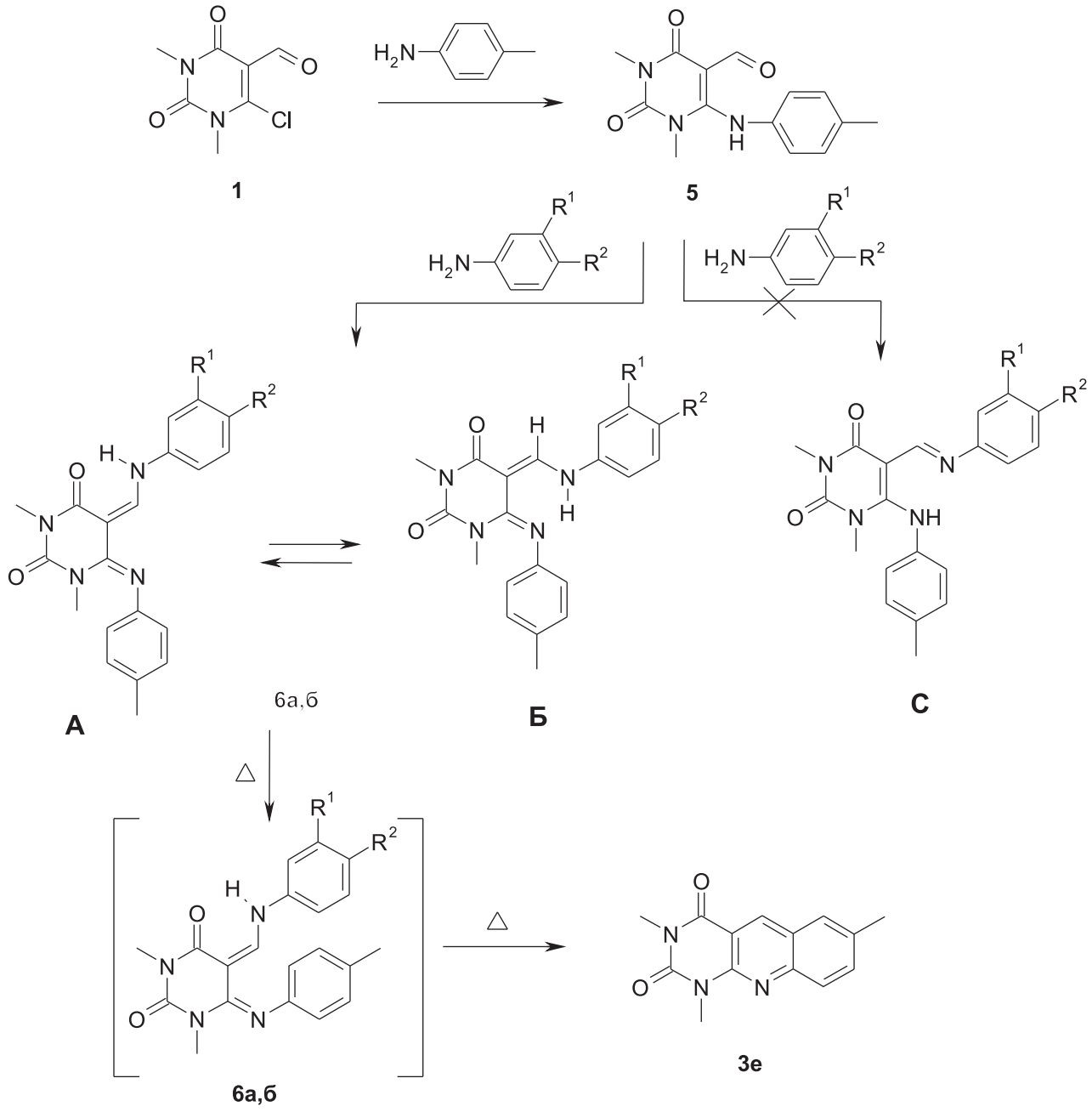
1,3-диметилурацилів і реагентів (диметилацеталь N,N-диметилформаміду, триетилортоформіат, дисульфід вуглецю, реагент Вільсмайєра) з наступним внутрішньомолекулярним циклоприєднанням [23-28]. Вихідною сполукою нами був обраний 1,3-диметил-5-форміл-6-хлороурацил **1**, при взаємодії якого з піперидином проходить нуклеофільне заміщення атома хлору з утворенням альдегіду **2**, який тривалий час зберігається при кімнатній температурі. Перетворення **2**→**3** відбувається при кип'ятінні 6-аміно-1,3-диметил-5-формілурацилу і ароматичних амінів у бутанолі з високими виходами. Слід зазначити, що 1,3-диметил-9-метокси-піримідо[4,5-*b*]хінолін-2,4(1*H*,3*H*)-діон **(3а)**, 1,3,7-триметилпіримідо[4,5-*b*]хінолін-2,4(1*H*,3*H*)-діон **(3е)** і 1,3-диметил-7-етилпіримідо[4,5-*b*]хінолін-2,4(1*H*,3*H*)-діон **(3ж)** були синтезовані раніше іншим методом [25, 28]. Нам вдалося також ввести в реакцію диметоксизаміщені ароматичні аміни і отримати продукти з високими виходами.

При взаємодії сполук **3а-и** зі спиртовим розчином гідроксиду натрію впродовж 1-5 год відбувається розщеплення піримідинового циклу і утворення амідів 2-(метиламіно)хінолін-3-карбонових кислот **4а-и** (схема 1).

Особливу увагу привертає реакція альдегіду **2** з мета-заміщеними ароматичними амінами, оскільки в процесі циклоприєднання утворюється суміш двох речовин – 6- та 8-заміщених 1,3-диметилпіримідо[4,5-*b*]хінолін-2,4(1*H*,3*H*)-діонів **3к-н** (схема 2). Про це свідчать дані спектрів ЯМР ^{1}H та ЯМР ^{13}C. Отримати сполуки в індивідуальному стані нам не вдалося, тому суміш продуктів була введена в реакцію гідролітичного розщеплення піримідинового циклу з наступним хроматографічним розділенням на колонці із силікагелем (елюент – гексан-етилацетат 1:1). При цьому з кількісним виходом були отримані сполуки **4к-н**.

Слід відмітити, що представлена нами реакція істотно доповнює описані в літературі методи отримання N-заміщених амідів 2-амінохінолін-3-карбонових кислот, серед яких знайдені речовини з вираженою біологічною активністю. Разом з тим вона є перспективним методом отримання нових представників хінолінів з метиламідними та метиламінозамісниками.

Для дослідження реакції отримання 1,3-диметилпіримідо[4,5-*b*]хінолін-2,4(1*H*,3*H*)-діонів нами були проведені наступні перетворення, представлені на схемі 3. Так, при взаємодії 1,3-диметил-5-форміл-6-хлороурацилу **1** з *n*-толуїдином проходить заміщення атома хлору з утворенням альдегіду **5**, який при нагріванні в бензолі з ароматичними амінами перетворюється на іміни **6а, б**.



R^1 = H, R^2 = Me (а); R^1 = CF_3, R^2 = H (б).

Схема 3

Зареєстровані спектри ЯМР ^{1}H та ^{13}C сполук **6** виявились досить складними для однозначної інтерпретації, оскільки в розчині продукт реакції може існувати у вигляді трьох таутомерних форм **А, Б, С.** Застосування методів гомо- і гетероядерної кореляційної спектроскопії ЯМР (NOESY, COSY, HSQC, HMBC) дозволило достовірно встановити таутомерні форми сполук **6**. Знайдені при цьому найважливіші кореляції, на основі яких і були зроблені віднесення сигналів, показані на рис. та в табл. 1.

Спін-спінова взаємодія CH-NH (7.3 і 9.7 Гц), а також крос-піки в спектрах NOESY сполуки **6а** 11.89↔6.38, 7.95, 11.89↔13.64, 11.89↔8.83, 11.89↔7.08 однозначно доводять, що сполука знаходиться в динамічній рівновазі і має в своєму складі (5*Z*,6*Z*)-1,3-диметил-5-{[(4-метилфеніл)аміно]метилено}-6-[(4-метилфеніл)іміно]дигідропіримідин-2,4(1*H*,3*H*)-діон (див. рис.). Таутомерної форми **С** при цьому не було встановлено. При нагріванні до ~120°С можлива аміно-імінна прототропія, яка полегшує син-анти-таутомерію в положенні 6 піримідинового циклу, що приводить до утворення сполуки, здатної до циклізації в продукт **3е** з відщепленням *п*-толуїдину чи *м*-(трифторометил)аніліну.

Склад і будова синтезованих сполук підтверджені результатами елементного аналізу, а також

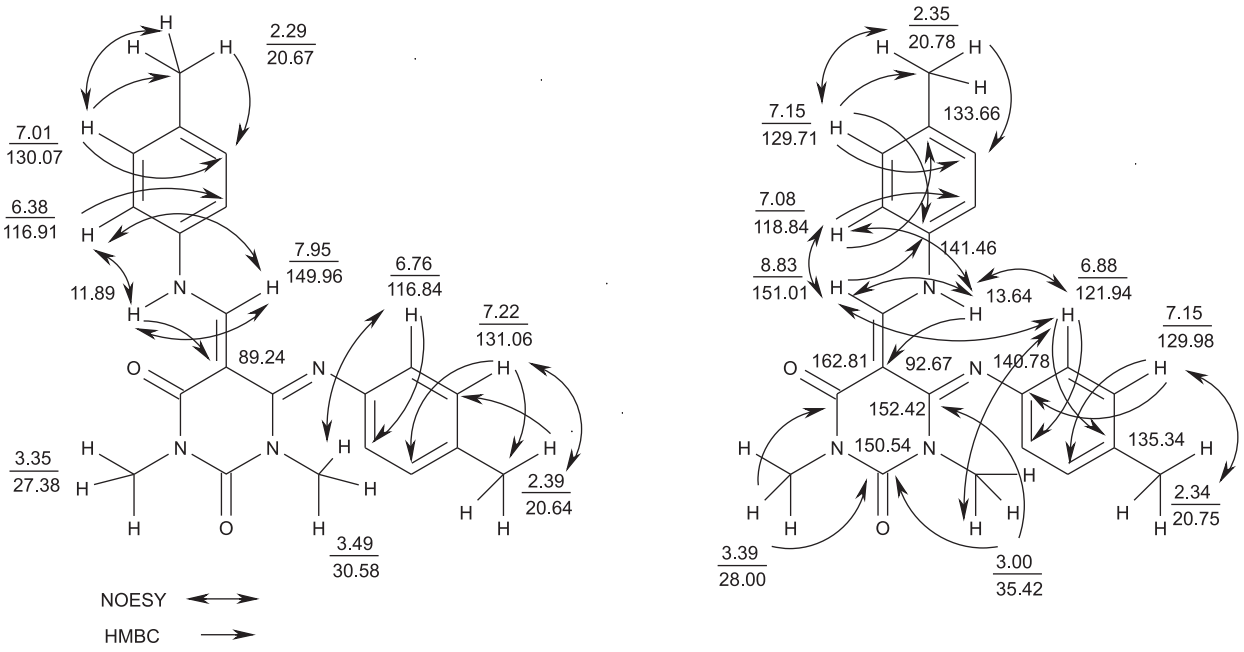


Рис. Основні кореляції (показані стрілками), віднесення сигналів (м.ч.) у спектрах ЯМР ^{1}H і ^{13}C сполуки **6а**.

даними хроматомас-спектрометрії та ЯМР спектроскопії.

Експериментальна частина

Спектри ЯМР ^{1}H та ^{13}C синтезованих сполук отримані на спектрометрі Bruker Avance 500 (500 і 125 МГц відповідно) в розчині $CDCl_3$ чи ДМСО-d_6 (для сполук **3д**, **3ж-и**), внутрішній стандарт – ТМС. Хроматомас-спектри одержані на приладі Agilent 1100 LC/MSD SL, метод іонізації – хімічна іонізація при атмосферному тиску (APCI), діапазон сканування – m/z 80-1000.

Таблиця 1

Перелік кореляцій, знайдених COSY, NOESY, HSQC, HMBC сполуки **6а**

^{1}H, δ	^{1}H, δ		^{13}C, δ	
	COSY	NOESY	HSQC	HMBC
2.29	7.01	7.01, 7.15, 2.35	20.67	130.07
7.01	2.29, 6.38	2.29, 2.35	130.07	130.07, 20.67
6.38	7.01	11.89, 7.95, 8.83, 13.64	116.91	116.91
11.89	7.95	6.38, 7.95, 13.64, 8.83, 7.08	–	89.24
7.95	11.89	6.38, 11.89, 13.64, 8.83, 7.08, 6.76	149.96	89.24
6.76	7.22	13.64, 8.83, 7.95, 3.49, 3.00	116.84	116.84
7.22	6.76	2.39	131.06	131.06, 20.64
2.39	2.39	7.22, 7.15	20.64	131.06
3.49	–	6.76, 3.00, 6.88	30.58	–
3.35	–	3.39	27.38	–
2.35	7.15	7.15, 7.01, 2.29	20.78	129.71
7.15	7.08, 6.88, 2.35, 2.34	2.35, 2.34,	129.71, 129.98	129.71, 141.46, 129.98, 140.78, 20.75
7.08	7.15	8.83, 13.64, 7.95, 11.89	118.84	118.84, 133.66
13.64	8.83	8.83, 7.08, 6.88, 6.38, 7.95, 11.89, 6.76	–	92.67
8.83	13.64	13.64, 7.08, 6.88, 6.38, 7.95, 11.89, 6.76	151.01	141.46
6.88	7.15	13.64, 8.83, 3.00, 3.49	121.94	121.94, 135.34
2.34	7.15	7.15, 2.39	20.75	129.98
3.00	–	6.88, 3.49, 6.76	35.42	152.42, 150.54
3.39	–	3.35	28.00	150.54, 162.81

Таблиця 2

Виходи, температури плавлення, мас-спектри та результати елементного аналізу синтезованих сполук **2**, **3а-и**, **4а-н**

Сполука	Вихід, %	Т. пл., °С	$[M+1]^+$	Знайдено, %			Формула	Вирахувано, %		
				C	H	N		C	H	N
2	67	104-106	252	57.42	6.70	16.74	$C_{12}H_{17}N_3O_3$	57.36	6.82	16.72
3а	54	244-245	272	61.95	4.78	15.54	$C_{14}H_{13}N_3O_3$	61.99	4.83	15.49
3б	61	211-213	260	60.27	4.07	16.29	$C_{13}H_{10}FN_3O_2$	60.23	3.89	16.21
3в	58	209-210	310	54.90	3.21	13.65	$C_{13}H_{10}F_3N_3O_2$	54.38	3.26	13.59
3г	73	284-286	302	60.02	5.12	13.98	$C_{15}H_{15}N_3O_4$	59.80	5.02	13.95
3д	81	277-278	302	59.71	4.97	14.04	$C_{15}H_{15}N_3O_4$	59.80	5.02	13.95
3е	70	226-228	256	65.91	5.18	16.54	$C_{14}H_{13}N_3O_2$	65.87	5.13	16.46
3ж	69	206-208	270	66.78	5.90	15.83	$C_{15}H_{15}N_3O_2$	66.90	5.61	15.60
3з	67	174-175	298	68.75	6.55	14.18	$C_{17}H_{19}N_3O_2$	68.67	6.44	14.13
3и	75	249-251	286	63.22	5.41	14.85	$C_{15}H_{15}N_3O_3$	63.15	5.30	14.73
4а	58	215-216	246	63.78	5.98	17.30	$C_{13}H_{15}N_3O_2$	63.66	6.16	17.13
4б	76	120-121	234	61.54	5.12	17.65	$C_{12}H_{12}FN_3O$	61.79	5.19	18.02
4в	45	201-202	284	55.01	4.34	14.96	$C_{13}H_{12}F_3N_3O$	55.13	4.27	14.83
4г	72	268-270	276	61.18	6.34	15.48	$C_{14}H_{17}N_3O_3$	61.08	6.22	15.26
4д	67	239-240	276	60.90	6.30	15.12	$C_{14}H_{17}N_3O_3$	61.08	6.22	15.26
4е	65	182-184	230	68.21	6.67	18.56	$C_{13}H_{15}N_3O$	68.10	6.59	18.33
4ж	62	219-220	244	69.18	6.79	17.35	$C_{14}H_{17}N_3O$	69.11	7.04	17.27
4з	89	98-100	272	70.57	7.67	15.51	$C_{16}H_{21}N_3O$	70.82	7.80	15.48
4и	66	156-158	260	64.97	6.78	16.53	$C_{14}H_{17}N_3O_2$	64.85	6.61	16.20
4к	44	123-125	246	63.58	5.99	17.00	$C_{13}H_{15}N_3O_2$	63.66	6.16	17.13
4л	48	119-121	234	61.64	5.29	17.55	$C_{12}H_{12}FN_3O$	61.79	5.19	18.02
4м	52	182-183	246	63.61	6.32	17.25	$C_{13}H_{15}N_3O_2$	63.66	6.16	17.13
4н	49	173-175	234	61.87	5.16	18.39	$C_{12}H_{12}FN_3O$	61.79	5.19	18.02
5	79	167-169	274	61.78	5.90	15.89	$C_{14}H_{15}N_3O_3$	61.53	5.53	15.38
6а	69	118-120 (розкл.)	363	70.02	6.15	15.74	$C_{21}H_{22}N_4O_2$	69.59	6.12	15.46
6б	74	120-121 (розкл.)	417	60.64	4.76	13.57	$C_{21}H_{19}F_3N_4O_2$	60.57	4.60	13.45

Контроль за перебігом реакції здійснювався методом тонкошарової хроматографії на пластинках Silufol, UV-254.

Фізико-хімічні і спектральні характеристики синтезованих сполук наведені в табл. 1-4.

1,3-Диметил-6-піперидин-1-іл-5-формілурацил (2). До суспензії 2,0 г (10 ммоль) альдегіду **1** в 15 мл етанолу при перемішуванні і охолодженні до 5-10°С додавали 1 мл піперидину. Реакційну суміш витримували впродовж 30 хв при 20-25°С. Осад, що випав, відфільтровували і очищали перекристалізацією із 2-пропанолу.

1,3-Диметилпіримідо[4,5-*b*]хінолін-2,4-(1*H*,3*H*)-діони (3а-и). Суміш 10 ммоль альдегіду **2** та 20 ммоль відповідного ароматичного аміну в 20 мл бутанолу кип'ятили впродовж 1-5 год (контроль ТШХ). Осад, що випав при охолодженні, відфільтровували і очищали перекристалізацією із ДМФА.

1,3,7-Триметилпіримідо[4,5-*b*]хінолін-2,4-(1*H*,3*H*)-діон (3е) *Метод Б.* 1 ммоль однієї із сполук **6а,б** нагрівали при 120°С до розплаву, залишали при тій же температурі ще на 15 хв. Охолоджений залишок обробляли 10 мл етанолу. Осад відфільтровували. Вихід – 90% (зі сполуки **6а**), 86% (зі сполуки **6б**). Проба змішування зразків, отриманих різними методами, не давала депресії температури плавлення.

***N*-Метил-2-(метиламіно)хінолін-3-карбоксаміди (4а-н).** Суміш 10 ммоль однієї із сполук **3а-и** і 20 ммоль гідроксиду натрію в 20 мл водного етанолу (1:1) кип'ятили впродовж 1-5 год (контроль ТШХ). Охолоджували, додавали 10 мл води, осад, що випав, відфільтровували, перекристалізовували із 2-пропанолу.

***N*-Метил-2-(метиламіно)-5-метокси(фторо)хінолін-3-карбоксаміди (4м, н) та *N*-метил-2-(метиламіно)-7-метокси(фторо)хінолін-3-**

Таблиця 3

ЯМР ^{1}H спектри сполук **2**, **3а-и**, **4а-н**, **5**

Сполука	Спектри ЯМР ^{1}H, $CDCl_3$, δ, м.ч.(*J*, Гц)
2	1.69-1.74 (6H, м, $3CH_2$), 3.11-3.18 (4H, м, $2CH_2$), 3.31 (3H, с, NCH_3), 3.44 (3H, с, NCH_3), 9.91 (1H, с, CH=O)
3а	3.51 (3H, с, NCH_3), 3.86 (3H, с, NCH_3), 4.08 (1H, с, OCH_3), 7.18 (1H, д, J = 7.5 Гц, CH), 7.44 (1H, т, J = 7.5 Гц, CH), 7.52 (1H, д, J = 7.5 Гц, CH), 8.98 (1H, с, H^5)
3б	3.57 (3H, с, NCH_3), 3.89 (3H, с, NCH_3), 7.49 (1H, м, CH), 7.56 (1H, т, J = 8.0 Гц, CH), 7.77 (1H, д, J = 8.0 Гц, CH), 9.07 (1H, с, H^5)
3в	3.55 (3H, с, NCH_3), 3.84 (3H, с, NCH_3), 7.69 (1H, д, J = 8.5 Гц, CH), 8.08 (1H, д, J = 8.5 Гц, CH), 8.32 (1H, с, CH), 9.07 (1H, с, H^5)
3г	3.35 (3H, с, NCH_3), 3.65 (3H, с, NCH_3), 3.92 (3H, с, OCH_3), 3.99 (3H, с, OCH_3), 7.28 (1H, с, CH), 7.52 (1H, с, CH), 8.81 (1H, с, H^5)
3д	3.36 (3H, с, NCH_3), 3.66 (3H, с, NCH_3), 3.90 (3H, с, OCH_3), 3.99 (3H, с, OCH_3), 6.94 (1H, с, CH), 7.09 (1H, с, CH), 8.81 (1H, с, H^5)
3е	2.54 (3H, с, CH_3), 3.52 (3H, с, NCH_3), 3.81 (3H, с, NCH_3), 7.64-7.68 (2H, м, 2CH), 7.88 (1H, д, J = 8.5 Гц, CH), 8.90 (1H, с, H^5)
3ж	1.29 (3H, т, J = 7.5 Гц, $CH_2C\underline{H}_3$), 2.52 (2H, к, J = 7.5 Гц, $C\underline{H}_2CH_3$), 3.34 (3H, с, NCH_3), 3.64 (3H, с, NCH_3), 7.73 (1H, д, J = 8.5 Гц, CH), 7.83 (1H, д, J = 8.5 Гц, CH), 7.91 (1H, с, CH), 8.92 (1H, с, H^5)
3з	0.93 (3H, т, J = 7.5 Гц, CH_3), 1.32-1.39 (2H, м, CH_2), 1.59-1.71 (2H, м, CH_2), 2.68-2.76 (2H, м, CH_2), 3.33 (3H, с, NCH_3), 3.63 (3H, с, NCH_3), 7.71 (1H, д, J = 8.0 Гц, CH), 7.81 (1H, д, J = 8.0 Гц, CH), 7.89 (1H, с, CH), 8.91 (1H, с, H^5)
3и	1.43 (3H, т, J = 7.0 Гц, $OCH_2C\underline{H}_3$), 3.36 (3H, с, NCH_3), 3.67 (3H, с, NCH_3), 4.19 (2H, к, J = 7.0 Гц, $OC\underline{H}_2CH_3$), 7.52 (1H, д, J = 8.5 Гц, CH), 7.58 (1H, с, CH), 7.86 (1H, д, J = 8.5 Гц, CH), 8.95 (1H, с, H^5)
4а	3.02 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 3.18 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 4.04 (3H, с, OCH_3), 6.38 (1H, ш. с NH), 7.00 (1H, д, J = 7.5 Гц, CH), 7.12-7.16 (2H, м, CH), 7.74 (1H, ш. с NH), 7.94 (1H, с, H^4)
4б	2.95 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 3.06 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 6.53 (1H, ш. с NH), 6.98-7.03 (1H, м, CH), 7.17 (1H, д, J = 8.0 Гц, CH), 7.22-7.26 (1H, м, CH), 7.66 (1H, ш. с NH), 7.81 (1H, с, H^4)
4в	3.05 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 3.13 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 6.31 (1H, ш. с NH), 7.37 (1H, т, J = 8.0 Гц, CH), 7.65 (1H, д, J = 8.0 Гц, CH), 7.80 (1H, ш. с NH), 7.99 (1H, с, CH), 8.00 (1H, с, H^4)
4г	2.96 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 3.07 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 3.86 (3H, с, OCH_3), 3.98 (3H, с, OCH_3), 6.35 (1H, ш. с NH), 6.75 (1H, с, CH), 7.06 (1H,с, CH), 7.64 (1H, ш. с NH), 7.78 (1H, с, H^4)
4д	2.96 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 3.08 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 3.76 (3H, с, OCH_3), 3.96 (3H, с, OCH_3), 6.29 (1H, с, CH), 6.62 (2H, ш. с NH, CH), 7.38 (1H, ш. с NH), 7.71 (1H, с, H^4)
4е	2.43 (3H, с, CH_3), 3.00 (3H, д, J = 5.0 Гц, $NHC\underline{H}_3$), 3.11 (3H, д, J = 5.0 Гц, $NHC\underline{H}_3$), 6.36 (1H, ш. с NH), 7.41 (1H, д, J = 8.5 Гц, CH), 7.60-7.62 (2H, м, CH, NH), 7.86 (1H, с, H^4)
4ж	1.30 (3H, т, J = 7.5 Гц, $CH_2C\underline{H}_3$), 2.74 (2H, к, J = 7.5 Гц, $C\underline{H}_2CH_3$), 3.01 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 3.11 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 6.30 (1H, ш. с NH), 7.32 (1H, с, CH), 7.45 (1H, д, J = 8.5 Гц, CH), 7.60 (1H, ш. с NH), 7.64 (1H, д, J = 8.5 Гц, CH), 7.89 (1H, с, H^4)
4з	0.93 (3H, т, J = 7.5 Гц, CH_3), 1.32-1.39 (2H, м, CH_2), 1.59-1.66 (2H, м, CH_2), 2.65 (2H, т, J = 7.5 Гц, CH_2), 2.96 (3H, д, J = 5.0 Гц, $NHC\underline{H}_3$), 3.06 (3H, д, J = 5.0 Гц, $NHC\underline{H}_3$), 6.45 (1H, ш. с NH), 7.23 (1H, с, CH), 7.39 (1H, д, J = 8.5 Гц, CH), 7.56 (1H, ш. с NH), 7.59 (1H, д, J = 8.5 Гц, CH), 7.84 (1H, с, H^4)
4и	1.47 (3H, т, J = 7.5 Гц, $OCH_2C\underline{H}_3$), 3.03 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 3.11 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 4.08 (2H, к, J = 7.5 Гц, $OC\underline{H}_2CH_3$), 6.29 (1H, ш. с NH), 6.89 (1H, с, CH), 7.30 (1H, д, J = 8.5 Гц, CH), 7.46 (1H, ш. с NH), 7.65 (1H, д, J = 8.5 Гц, CH), 7.87 (1H, с, H^4)
4к	2.96 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 3.08 (3H, д, J = 4.5 Гц, $NHC\underline{H}_3$), 3.89 (3H, с, OCH_3), 6.35 (1H, ш. с NH), 6.78 (1H, д, J = 8.5 Гц, CH), 7.03 (1H, с, CH), 7.35 (1H, д, J = 8.5 Гц, CH), 7.82-7.87 (2H, м NH, H^4)
4л	2.99 (3H, д, J = 5.0 Гц, $NHC\underline{H}_3$), 3.08 (3H, д, J = 5.0 Гц, $NHC\underline{H}_3$), 6.33 (1H, ш. с NH), 6.89 (1H, т, J = 8.5 Гц, CH), 7.29 (1H, д, J = 8.5 Гц, CH), 7.44 (1H, т, J = 8.5 Гц, CH), 7.77 (1H, ш. с NH), 7.86 (1H, с, H^4)
4м	2.99 (3H, д, J = 5.0 Гц, $NHC\underline{H}_3$), 3.08 (3H, д, J = 5.0 Гц, $NHC\underline{H}_3$), 6.28 (1H, ш. с NH), 6.51 (1H, д, J = 8.0 Гц, CH), 7.26 (1H, д, J = 8.0 Гц, CH), 7.45 (1H, т, J = 8.0 Гц, CH), 7.86 (1H, ш. с NH), 8.38 (1H, с, H^4)
4н	3.00 (3H, д, J = 5.0 Гц, $NHC\underline{H}_3$), 3.08 (3H, д, J = 5.0 Гц, $NHC\underline{H}_3$), 6.42 (1H, ш. с NH), 6.80 (1H, т, J = 8.0 Гц, CH), 7.38-7.49 (2H, м, 2CH), 7.82 (1H, ш. с NH), 8.19 (1H, с, H^4)
5	2.40 (3H, с, CH_3), 3.04 (3H, с, NCH_3), 3.40 (3H, с, NCH_3), 7.02 (2H, д, J = 7.5 Гц, 2CH), 7.23 (2H, д, J = 7.5 Гц, 2CH), 10.03 (1H, с, CH=O), 12.37 (1H, с, NH)

Таблиця 4

ЯМР ^{13}C спектри сполук **2**, **3а-и**, **4а-н**, **5**

Сполука	Спектри ЯМР ^{13}C, $CDCl_3$, δ, м.ч.(*J*, Гц)
2	23.3 (CH_2), 25.2 ((CH_2)$_2$), 27.7 (NCH_3), 34.5 (NCH_3), 51.5 (N(CH_2)$_2$), 99.9 (C^5), 152.1 (C=O), 160.4 (C^6), 163.9 (C=O), 186.5 (CH=O)
3а	28.2 (NCH_3), 29.7 (NCH_3), 56.1 (OCH_3), 110.9 (C), 111. 2 (CH), 120.6 (CH), 125.6 (C), 125.9 (CH), 139.7 (CH), 141.4 (C), 147.6 (C), 151.5 (C), 154.3 (C=O), 161.2 (C=O)
3г	28.3 (NCH_3), 29.4 (NCH_3), 56.0 (OCH_3), 56.3 (OCH_3), 105.8 (CH), 106.3 (CH), 108.3 (C), 120.1 (C), 137.0 (CH), 147.4 (C), 147.6 (C), 149.3 (C), 151.4 (C), 155.6 (C=O), 161.6 (C=O)
3з	13.8 (CH_3), 22.1 (CH_2), 28.4 (NCH_3), 29.4 (NCH_3), 32.9 (CH_2), 35.1 (CH_2), 110.5 (C), 124.5 (C), 127.1 (CH), 127.7 (CH), 134.7 (CH), 139.2 (CH), 140.4 (C), 147.6 (C), 148.4 (C), 151.3 (C=O), 161.2 (C=O)
3и	14.5 (CH_3), 28.1 (NCH_3), 29.2 (NCH_3), 63.9 (CH_2), 106.1 (CH), 110.7 (C), 125.5 (C), 126.6 (CH), 129.5 (CH), 137.9 (CH), 145.8 (C), 146.8 (C), 151.5 (C), 156.4 (C=O), 161.5 (C=O)
4а	26.7 (CH_3), 27.9 (CH_3), 56.2 (OCH_3), 110.4 (CH), 115.9 (C), 120.4 (CH), 121.7 (CH), 122.2 (C), 136.0 (CH), 140.9 (C), 153.0 (C), 155.2 (C), 168.9 (C=O)
4г	26.6 (CH_3), 27.9 (CH_3), 55.7 (OCH_3), 55.8 (OCH_3), 105.8 (CH), 106.4 (CH), 112.6 (C), 115.1 (C), 134.3 (CH), 146.4 (C), 146.5 (C), 153.6 (C), 155.2 (C), 169.2 (C=O)
4д	26.3 (CH_3), 27.5 (CH_3), 54.9 (OCH_3), 55.8 (OCH_3), 97.9 (CH), 102.5 (CH), 115.9 (C), 121.6 (C), 134.7 (CH), 136.4 (C), 153.4 (C), 153.7 (C), 154.2 (C), 168.7 (C=O)
4ж	15.5 (CH_3), 26.7 (CH_2), 28.0 (CH_3), 28.4 (CH_3), 115.6 (C), 121.3 (C), 125.8 (CH), 125.9 (CH), 132.4 (CH), 135.6 (CH), 137.9 (C), 148.0 (C), 155.4 (C), 169.1 (C=O)
4з	13.7 (CH_3), 22.2 (CH_2), 26.4 (CH_3), 27.9 (CH_3), 33.4 (CH_2), 35.0 (CH_2), 115.5 (C), 121.2 (C), 125.7 (CH), 126.6 (CH), 132.5 (CH), 135.4 (CH), 136.4 (C), 147.8 (C), 155.3 (C), 168.9 (C=O)
4к	26.4 (CH_3), 27.7 (CH_3), 55.4 (CH_3), 104.8 (CH), 112.5 (C), 114.3 (CH), 115.9 (C), 129.0 (CH), 135.2 (CH), 151.3 (C), 156.3 (C), 162.5 (C), 168.9 (C=O)
4л	26.7 (CH_3), 27.8 (CH_3), 109.9 д (CH, J_{C-F} = 22.0 Гц), 111.8 д (CH, J_{C-F} = 22.0 Гц), 114.5 (C), 117.9 (C), 129.8 (CH), 135.3 (CH), 150.7 (C), 156.2 (C), 164.7 д (C, J_{C-F} = 237.5 Гц), 168.6 (C=O)
4н	26.7 (CH_3), 27.8 (CH_3), 105.6 д (CH, J_{C-F} = 20.0 Гц), 111.2 д (C, J_{C-F} = 15.5 Гц), 115.3 (C), 121.7 (CH), 128.6 (CH), 130.9 (CH), 150.2 (C), 156.1 (C), 156.9 д (C, J_{C-F} = 250.0 Гц), 168.5 (C=O)
5	20.9 (CH_3), 27.8 (NCH_3), 35.4 (NCH_3), 94.3 (C^5), 124.2 (2CH), 130.3 (2CH), 134.7 (C), 137.2 (C), 151.5 (C=O), 157.4 (C^6), 162.2 (C=O), 189.1 (CH=O)

карбоксаміди (4к, л) екстрагували з водного розчину етилацетатом, розчинник упарювали, залишок розділяли на колонці з силікагелем (елюент гексан-етилацетат 1:1).

1,3-Диметил-6-[(4-метилфеніл)аміно]-5-формілурацил (5). До суспензії 2.0 г (10 ммоль) альдегіду **1** в 15 мл етанолу при перемішуванні і охолодженні до 5-10°С додавали суміш 10 ммоль триетиламіну та 10 ммоль *n*-толуїдину. Реакційну суміш витримували протягом 1 год при 20-25°С. Осад, що випав, відфільтровували і очищали перекристалізацією із етанолу.

(5*Z*,6*Z*)-1,3-Диметил-5-[(ариламіно)метилено]-6-[(4-метилфеніл)іміно]-дигідропіримідин-2,4(1*H*,3*H*)-діони (6а, б). Суспензію 10 ммоль альдегіду **5** та 10 ммоль одного із анілінів в 20 мл бензолу кип'ятили протягом 1-2 год. Осад, що випав, відфільтровували і очищали перекристалізацією із ацетонітрилу.

Висновки

1. Знайдено зручний підхід до синтезу заміщених 2-(метиламіно)хінолін-3-карбоксамідів реакцією гідролітичного розщеплення піримідинового циклу в піримідо[4,5-*b*]хінолін-2,4(1*H*,3*H*)-діонах (5-деазаалоксазинах).

2. За допомогою методів гомо- і гетероядерної кореляційної спектроскопії ЯМР встановлено будову продуктів реакції конденсаціїї похідних 1,3-диметил-5-формілурацилу з ароматичними амінами.

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