Научно-практический журнал
ФАРМАЦИЯ И ФАРМАКОЛОГИЯ

оригинальная статья

DOI: 10.19163/2307-9266-2021-9-2-149-160

PHARMACOKINETIC PROPERTIES OF A NEW KAPPA-OPIOID ANALGESIC RU-1205 COMPOUND AT A SINGLE PERORAL ADMINISTRATION

A.A. Spasov[1], L.A. Smirnova[1,3], O.Yu. Grechko[1], N.V. Eliseeva[1], Yu.V. Lifanova[1], A.I. Rashchenko[1], O.N. Zhukovskaya[2], A.S. Morkovnik[2], V.A. Anisimova[2]

[1] Volgograd State Medical University
1, Pavshikh Bortsov Square, Volgograd, Russia 400131
[2] Scientific Research Institute of Physical and Organic Chemistry, Southern Federal University
Bldg. 2, 194, Stachki Ave., Rostov-on-Don, Russia, 344090
[3] Volgograd Medical Research Center;
1, Pavshikh Bortsov Square, Volgograd, Russia, 400131

E-mail: j_semenova_pharm@mail.ru

Received 21 Jan 2021 Accepted 24 Apr 2021

The aim of the study is the investigation of the pharmacokinetic properties of the RU-1205 compound, with previously identified kappa-agonistic and analgesic effects, at a single oral administration, as well as comparison of the relationship between its pharmacokinetic and analgesic properties.

Materials and methods. Pharmacokinetic parameters of RU-1205 after the oral administration at the dose of 50 mg/kg were investigated using the method of High Performance Liquid Chromatography with determination of the concentration of the compound according to the previously constructed calibration schedule. The indices of the area under the pharmacokinetic curve, clearance, half-life, residence time of the drug molecule in the body, a total (apparent) volume of distribution, as well as the indicator of absolute bioavailability, were calculated. The tissue distribution and excretion of RU-1205 were studied. Potential metabolites of RU-1205 were predicted using the PALLAS 3.00 program. The study of the analgesic activity was carried out on a model of central somatogenic pain with electricalstimulation, with the dynamics assessment of the voltage amplitude of the corresponding reaction of the «tail-flick» reflex.

Results. The compound under study is rapidly adsorbed from the gastrointestinal tract, reaching a maximum concentration by the end of the first hour of the study, and is determined in plasma within 12 hours. Its half-life is 17.7 hours. The absolute oral bioavailability is 37.3%. It was found out that the compound is withdrawn within 3–4 days. The main route of excretion is extrarenal. Biotransformation of a substance probably proceeds mainly with the formation of oxidized forms of the initial molecule by reactions of the first phase of metabolic transformation. The analgesic effect is long-lasting: it starts after 15 minutes and lasts for 12 hours with flattening of the curve by the 8th hour.

Conclusion. When administered orally, the test substance undergoes a long process of elimination, has the greatest tropism for the elimination organs and undergoes active biotransformation processes in the body of animals. As a result of it, active metabolic products with an analgesic activity are, possibly, formed.

Keywords: kappa agonists; opioid analgesics; benzimidazoles; pharmacokinetics; peroral route; bioavailability; excretion; tissue distribution

List of abbreviations: HPLC – High Performance Liquid Chromatography; RCF – relative centrifugal field; AUC – area under the curve; Kel – elimination constant; Cl – clearance; T1/2 – half-life of a drug; MRT – mean residence time; Vd – Volume of distribution; AVD – apparent volume of distribution; TA – tissue availability; ACN – acetonitrile; GIT – gastrointestinal tract; МАРК – mitogen-activated protein kinase

For citation: A.A. Spasov, L.A. Smirnova, O.Yu. Grechko, N.V. Eliseeva, Yu.V. Lifanova, A.I. Rashchenko, O.N. Zhukovskaya, A.S. Morkovnik, V.A. Anisimova. Pharmacokinetic properties of a new kappa-opioid analgesic RU-1205 compound at a single peroral administration. *Pharmacy & Pharmacology.* 2021;9(2):149-160. **DOI:** 10.19163/2307-9266-2021-9-2-149-160

Для цитирования: А.А. Спасов, Л.А. Смирнова, О.Ю. Гречко, Н.В. Елисеева, Ю.В. Лифанова, А.И. Ращенко, О.Н. Жуковская, А.С. Морковник, В.А. Анисимова. Фармакокинетические свойства нового каппа-опиоидного анальгетика – соединения RU-1205 при однократном пероральном введении. *Фармация и фармакология.* 2021;9(2):149-160. **DOI:** 10.19163/2307-9266-2021-9-2-149-160

RESEARCH ARTICLE

ISSN 2307-9266 e-ISSN 2413-2241

Scientific and Practical Journal
PHARMACY & PHARMACOLOGY

ФАРМАКОКИНЕТИЧЕСКИЕ СВОЙСТВА НОВОГО КАППА-ОПИОИДНОГО АНАЛЬГЕТИКА – СОЕДИНЕНИЯ RU-1205 ПРИ ОДНОКРАТНОМ ПЕРОРАЛЬНОМ ВВЕДЕНИИ

А.А. Спасов[1], Л.А. Смирнова[1,3], О.Ю. Гречко[1], Н.В. Елисеева[1], Ю.В. Лифанова[1], А.И. Ращенко[1], О.Н. Жуковская[2], А.С. Морковник[2], В.А. Анисимова[2]

[1] Федеральное государственное бюджетное образовательное учреждение высшего образования «Волгоградский государственный медицинский университет» Министерства здравоохранения Российской Федерации
400131, Россия, г. Волгоград, площадь Павших Борцов, д. 1
[2] Научно-исследовательский институт физической и органической химии федерального государственного автономного образовательного учреждения высшего образования «Южный федеральный университет»
344090, г. Ростов-на-Дону, пр. Стачки, 194/2
[3] Государственное бюджетное учреждение «Волгоградский медицинский научный центр»
400131, г. Волгоград, площадь Павших Борцов, д. 1

E-mail: j_semenova_pharm@mail.ru

Получено 21.01.2021 Принята к печати 24.04.2021

Цель. Исследование фармакокинетических свойств соединения RU-1205, с ранее выявленным каппа-агонистическим и анальгетическим действием, при однократном пероральном введении, а также сопоставление взаимосвязи между его фармакокинетическими и обезболивающими свойствами.
Материалы и методы. Фармакокинетические параметры RU-1205 после перорального введения в дозе 50 мг/кг исследовали с помощью метода высокоэффективной жидкостной хроматографии с определением концентрации соединения по предварительно построенному калибровочному графику. Рассчитывали показатели площади под фармакокинетической кривой, клиренса, периода полувыведения, время пребывания в организме молекулы препарата, общего (кажущегося) объём распределения, а также показатель абсолютной биодоступности. Изучали тканевое распределение и экскрецию RU-1205. Осуществляли прогноз возможных метаболитов соединения RU-1205 с помощью программы «PALLAS 3.00». Исследование анальгетической активности проводили на модели центральной соматогенной боли при электрическом раздражении с оценкой динамики амплитуды напряжения соответствующей реакции «отдергивания хвоста».
Результаты. Изучаемое соединение быстро адсорбируется из желудочно-кишечного тракта с достижением максимальной концентрации к концу первого часа исследования и определяется в плазме в течение 12 часов. Период полувыведения составляет 17,7 ч. Абсолютная биодоступность при пероральном пути – 37,3%. Установлено, что соединение выводится за 3–4 дня. Основной путь выведения внепочечный. Биотрансформация вещества вероятно протекает в основном с образованием окисленных форм исходной молекулы по реакциям первой фазы метаболической трансформации. Анальгетическое действие продолжительное: начинается через 15 минут и сохраняется в течении 12 часов с выходом на плато к 8 часу.
Заключение. Исследуемое вещество подвергается длительному процессу элиминации при пероральном введении, имеет наибольшую тропность к органам элиминации и подвергается активным процессам биотрансформации в организме животных, в результате которых, возможно, образуются активные продукты метаболизма с анальгетической активностью.
Ключевые слова: каппа-агонисты; опиоидные анальгетики; бензимидазолы; фармакокинетика; пероральный путь; биодоступность; экскреция; тканевое распределение
Список сокращений: ВЭЖХ – высокоэффективная жидкостная хроматография; AUC – площадь под фармакокинетической кривой; Kel – константа элиминации; Cl – клиренс; $T_{1/2}$ – продолжительность периода полувыведения; MRT – среднее время пребывания в организме молекулы; Vd – общий (кажущийся) объём распределения; fT – тканевая доступность; ACN – ацетонитрил; ЖКТ – желудочно-кишечный тракт; МАРК – митоген-активируемая протеинкиназа.

INTRODUCTION

For many decades, opioid analgesics continue to be the mainstay of the pharmacotherapy of severe pain syndromes. However, the global opioid crisis requires a change in the established generally accepted practice of opioids clinical use with the need to replace some of the traditionally used narcotic analgesics with a narrow therapeutic index, a pronounced narcogenic potential, and non-optimal pharmacokinetics [1, 2]. Nonselective agonists of opioid receptors (for example, morphine), especially in injections, have a rather high risk of non-medical use due to their pronounced narcogenic potential (due to the activation of predominantly μ-opioid receptors). This significantly complicates their regulatory accessibility, complicates the work of medical personnel due to complex accounting rules for narcotic analgesics, in-

DOI: 10.19163/2307-9266-2021-9-2-149-160

creasing the degree of control in accordance with the Decree of the Government of the Russian Federation No. 681 dated June 30, 1998 (as amended on 03.12.2020), and thereby significantly limits them clinical use, not fully meeting the patients' need for strong analgesics approved for use [3].

To solve the problem of narcogenicity of opioid analgesics, several directions have been proposed. One of them is the search and development of new opioid analgesics with a safer profile of a receptor activity due to the selectivity of the drug's action on various subtypes of opioid receptors, which will ensure an increase in the effectiveness of the analgesia performed together with minimization of side effects. [4.5]. Partial agonists and mixed agonists-antagonists of various subpopulations of opioid receptors (buprenorphine, butorphanol, nalbuphine), developed to increase the selectivity of the receptor profile, have found only a limited use in clinical practice, since they had retained a number of negative properties of traditional opiates, in particular narcogenicity [6].

The study of the analgesic properties of RU-1205 in experimental nociceptive models showed that in the analgesic activity, the studied compound is superior to morphine and butorphanol [7]. Unlike the reference drugs, it does not cause respiratory depression and does not possess pharmacological properties that can be regarded as specific predictors of substance capacity to form physical dependence, cause aversion or addiction [8,9]. According to the modern concept of therapy for a severe chronic pain, non-invasive dosage forms of opioid analgesics are recognized as the most effective and safe drugs that provide the best life quality for patients (except the final stages of the disease). One of the important advantages of the test substance is its effectiveness in various administration modes, including an oral administration. This opens up prospects for the creation of an effective oral tablet dosage form on its basis, in contrast to butorphanol, which undergoes intensive metabolism in the liver and is used in medical practice only in the form of an injection solution [10].

At the stage of creating new original drugs, one of the necessary stages of preclinical studies is the research of the pharmacokinetic parameters of the compound being developed. These parameters will make it possible to solve the whole range of the applied issues, such as studying the degree and the rate of adsorption, biotransformation, permeability of drugs through tissue barriers and distribution between blood and peripheral tissues, excretion of the studied drugs, as well as assessing bioavailability during an extravascular administration of a drug [11].

THE AIM of the study is the investigation of the pharmacokinetic properties of the RU-1205 compound at a single oral administration, as well as comparison of the relationship between its pharmacokinetic and analgesic properties.

MATERIALS AND METHODS

Experimental animals

The studies were carried out on sexually mature male rats weighing 200-230 g, in the amount of 74 individuals, obtained from the Rappolovo laboratory animal nursery of the Russian Academy of Medical Sciences (St. Petersburg, Russia). Before the start of the experiment, the animals were subjected to the adaptation quarantine for 14 days in the vivarium of the Department of Pharmacology and Bioinformatics of Volgograd State Medical University (the Ministry of Health of Russia). The rats had a round-the-clock access to feeding troughs and drinkers *ad libitum*, and were kept in standardized vivarium conditions (Decree of August 29, 2014 No. 51 "On the approval of SP 2.2.1.3218-14" Sanitary and epidemiological requirements for the device, equipment and maintenance of experimental biological clinics (vivariums) "; Directive of the European Parliament and the Council of the European Union 2010/63 / EU of September 22, 2010" On the protection of animals used for scientific purposes"). The animals were kept in groups of 5 individuals in a controlled combined light regime (12/12 h) and the temperature of 20-22°C. 12 hours before the experiment, the animals were deprived of food, but there was a free access to water. The experiments were approved by the Regional Research Ethics Committee of Volgograd Region (registration number IRB 00005839 IORG 0004900 (OHRP), protocol No. 2077-2018 dated October 30, 2018).

Test substances

Compound RU-1205 is (9-(2-morpholinoethyl)-2-(4-fluorophenyl)imidazo[1,2-a]-benzimidazole dihydrochloride) (Fig. 1) in the form of a substance synthesized at the Scientific Research Institute of Physical and Organic Chemistry of Rostov-on-Don Southern Federal University (Pat. RF No. 2 413 512 C1 dated July 29, 2009). The purity of the compound was at least 99.46%.

Study design

At the first stage of the investigation, the pharmacokinetic properties of RU-1205 were studied at the dose of 50 μg/kg at a single intragastric administration by method of High Performance Liquid Chromatography. The animals were randomly divided into 7 groups – a control group that received a 0.9% sodium chloride solution in the volume of 100 μl per 100 g of animal weight, and 6 groups that received the RU-1205 compound at the dose of 50 μg/kg (n=6). For the intragastric administration, atraumatic probe No. 14 (Kent Scientific, USA) was used. In order to determine the background peaks before the experiment, the blood was taken from all 42 animals, followed by obtaining plasma samples in the volume of 1.5–2 ml. After the administration of the RU-1205 compound in 30 minutes, 1, 2, 4, 8, 12 hours,

the animals were decapitated using a guillotine (OpenScience, Russia), followed by blood and organ sampling. Decapitation and material sampling from the control group animals were carried out 20 minutes after the injection of the solvent. To obtain plasma, the blood samples were stabilized with a 5% aqueous sodium citrate solution (Polisintez, Russia) (pH 6.0), followed by centrifugation in a 15 min. mode at 3000 rpm, RCF = 604 g (ELMI, Latvia). For precipitation of plasma proteins and extraction of the test substance, acetonitrile was added to the blood samples in a 1:1 ratio. The organs under study (brain, liver, kidneys, heart, lungs, omentum, and muscle tissue) were subjected to grinding and homogenization (Silent Crusher, Germany) to obtain a 20% aqueous homogenate.

The study of tissue bioavailability is an integral part of pharmacokinetic research of new compounds. After obtaining the results, it is possible to judge the intensity of the substances distribution between the peripheral tissues and the target organ (for the RU-1205 compound, the target organ is the brain). When studying the tissue availability of RU-1205, the concentration of the compound was investigated in the following organs: the brain, the elimination organs (liver, kidneys), heart, lungs in especially vascularized tissues, moderately vascularized tissues (muscles, the quadriceps femoris muscle was selected for the experiment), omentum (in weakly vascularized tissues) [12].

The quantitative determination of RU-1205 in blood plasma and organ homogenates was studied according to the previously developed method [13] using High Performance Liquid Chromatography (HPLC) with a diode array detection on a liquid chromatograph (Shimadzu, Japan). The determination was carried out with High Performance Liquid Chromatography System consisting of a SUPELCOSIL LC-18 chromatographic column (5 µm 100×4.6 mm) with a diode-matrix ultraviolet detector (thermostatting temperature of 50°C). To prepare the mobile phase, acetonitrile (UF210) (Russia) was used in a 1:1 ratio with a buffer system (monosubstituted potassium phosphate 50 mM, pH = 5.0). The detection wavelength was 205 nm, and the flow rate of the mobile phase was 1 ml/min. The concentration of the RU-1205 compound in the samples was determined according to the previously constructed calibration graph. For this, the dependence of standard concentrations (0.5; 1; 5; 10; 25 µg/ml) on the areas of chromatographic peaks was plotted using the method of absolute standards. The regression coefficient (R^2) was 0.998.

To assess the pharmacokinetic properties of RU-1205 using Microsoft Office Excel 2007 software (USA), the following pharmacokinetic parameters were calculated:

1. AUC (Area Under the Curve) – the area under the pharmacokinetic curve "concentration – time" (by a model-independent method of statistical moments [14]);
2. Kel – elimination constant;
3. Cl – clearance;
4. T1/2 – the duration of the half-life;
5. MRT is the mean residence time of the RU-1205 molecule in the body;
6. Vd – total (apparent) volume of distribution.

The evaluation of the penetration intensity of the RU-1205 compound into various organs and tissues was carried out using the tissue availability parameter (fT), which is the ratio of the organ / tissue AUC value to the blood plasma AUC value.

At the second stage of the study, the excretion of the RU-1205 compound was studied by HPLC in urine and feces samples at a single intragastric administration. The experiments were carried out on 20 sexually mature male rats, randomly divided into control and experimental groups (n=10). The experimental group received a single injection of the RU-1205 compound at the dose of 50 mg/kg intragastrically with an atraumatic probe, the animals from the control group received a solvent – a 0.9% sodium chloride solution. After 24, 48, 72 and 96 hours, the samples were taken in the Thermoplast metabolic chambers (Italy). The samples were subjected to the sample preparation and analysis by HPLC as described above.

At the third stage, the dynamics of the analgesic activity was studied depending on the concentration of RU-1205 in the blood plasma after a single intragastric administration in the electric stimulation test of the tail head. The studies were carried out on 12 sexually mature male rats (n=6). The animals of the control group were injected once intragastrically with the RU-1205 compound at the dose of 5 mg/kg with an atraumatic probe (0.9% sodium chloride solution). For 12 hours, the analgesic activity was studied in the electric stimulation test of the tail head when stimulation was applied through subcutaneous electrodes (rectangular pulses with a frequency of 100 Hz, the duration of 10 ms, the stimulation duration of 1 sec) (Laboratory electro-stimulator ESL-2, Russia) with gradual sequential increase in voltage [15, 16]. The value of the pain threshold (before the manifestation of the spinal "*tail-flic* " reflex) was assessed as an indicator of tension, expressed in volts. The presence of analgesic properties of the RU-1205 compound was judged on the basis of the voltage amplitude changes compared with the values of the control animals.

The PALLAS 3.00 program (CompuDrug Chemistry Ltd.) was used to predict possible metabolites of RU-1205.

Statistical processing of results

Statistical processing of the obtained data was carried out using Microsoft Office Excel 2007 Software (USA).

DOI: 10.19163/2307-9266-2021-9-2-149-160

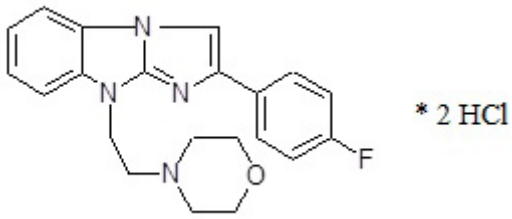
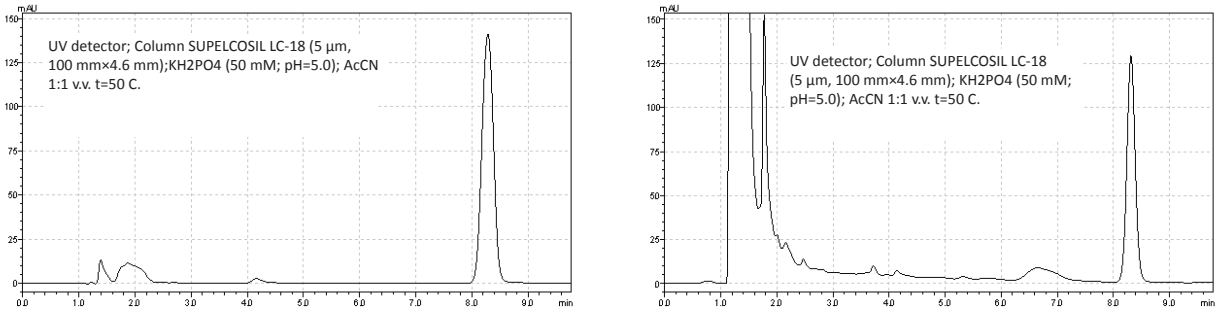


Figure 1 – Structural formula of RU-1205 compound



A **B**

Figure 2 – Chromatogram of substance of RU-1205 compound at concentration of 5 µg/ml in aqueous solution (A) and biological material (B). Retention time – 8.00–8.83 min

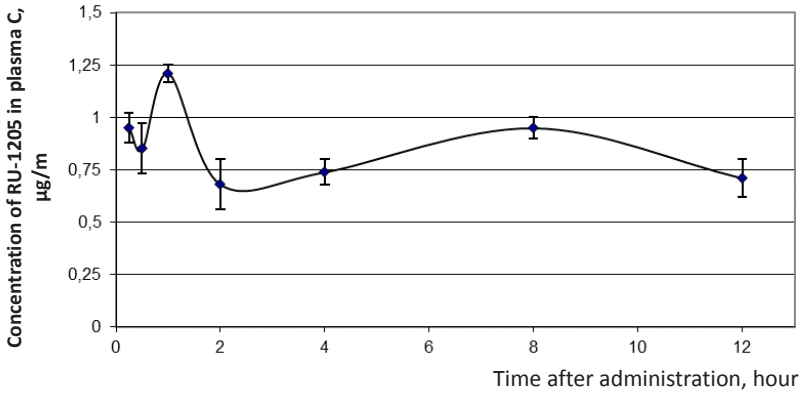
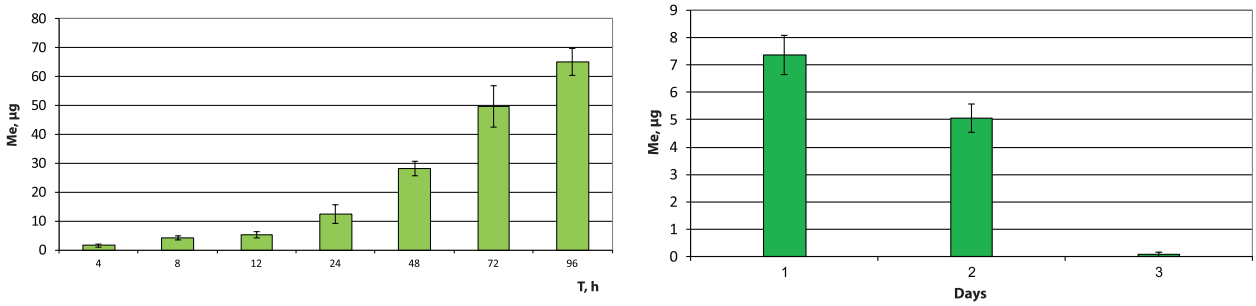


Figure 3 – Kinetic curve of RU-1205 compound in rat blood plasma
Note: administration – oral, dose – 50 mg/kg



A **B**

Figure 4 – Cumulative excretion of compound RU-1205 through the kidneys (A) and gastrointestinal tract (B) in intragastric administration
Note: on the abscissa – time, h; on the ordinate – amount of RU-1205, µg

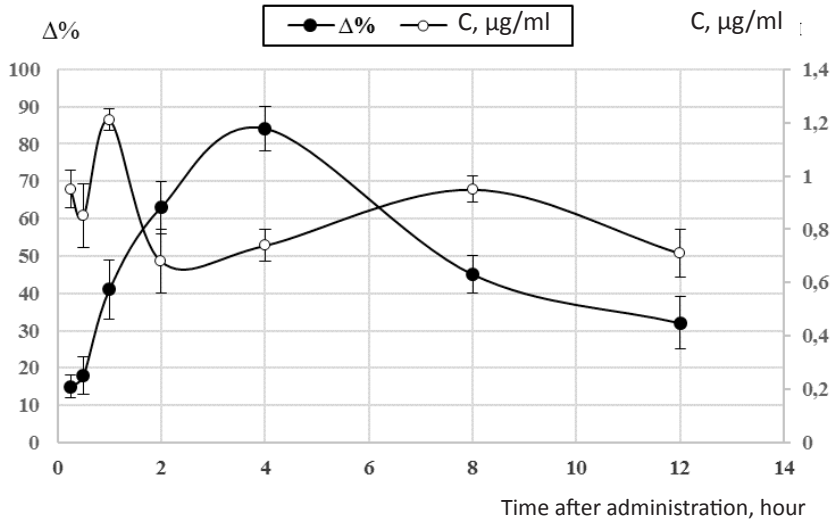
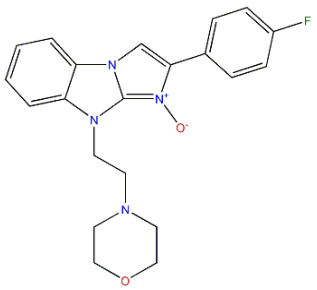
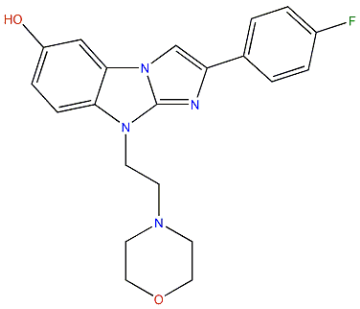
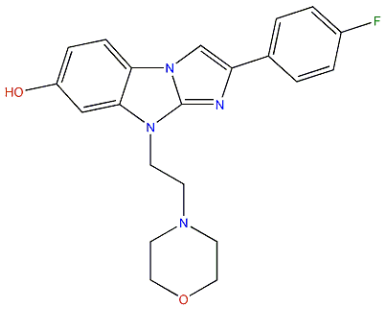
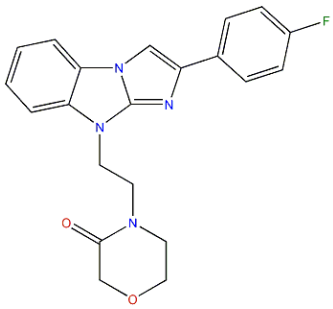
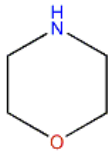
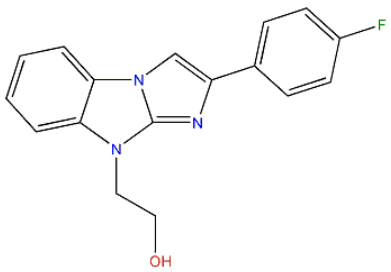
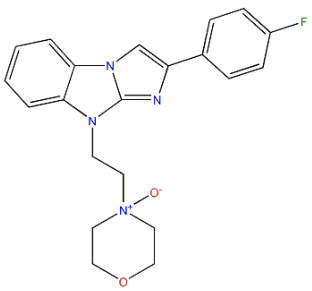
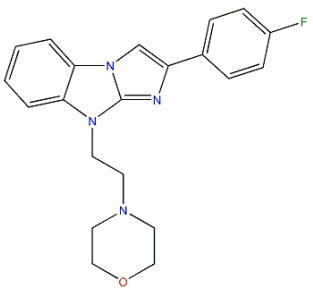


Figure 5 – Dependence of the dynamics of RU-1205 antinociceptive activity on the concentration of the compound in blood plasma and time at intragastric administration

Note: on the abscissa – time (hours); on the ordinate: on the left – pain threshold shift (Δ% in relation to the control), on the right – compound concentration in blood plasma (μg/ml)

Table 1 – Projection of possible metabolites of RU-1205 compound *in silico* (program "PALLAS 3.00")

A	B	C
		
D	**E**	**F**
		
G	**Structure of RU-1205 compound**	
		

DOI: 10.19163/2307-9266-2021-9-2-149-160

RESULTS

Using the developed method of quantitative determination, chromatograms of standard aqueous and plasma solutions of RU-1205 were obtained (Fig. 2).

The pharmacokinetic curve of the concentration changes in the substance of the RU-1205 compound in the blood plasma after the oral administration is shown in Fig. 3. The figure shows that the kinetic curve has a two-phase nature of the dynamics of concentration changes. The first phase of increasing the concentration characterizes the process of the substance absorption. The rapid stage of concentration increasing begins from the 30-th minute, and the maximum concentration of the compound in the plasma (C_{max} = 1.05 μg/ml) is already reached by the 60-th minute of the study, which indicates the rapid adsorption of the compound RU-1205 from the gastrointestinal tract (Fig. 3). The second part of the pharmacokinetic curve characterizes the elimination of the substance of the compound from the blood plasma, initially with a sharp decrease in the concentration during the second hour of the study and a subsequent gradual decrease within 12 hours, which indicates a long elimination process. The area under the pharmacokinetic curve was 27.56 μg*hour/ml. The long-term nature of the elimination of the RU-1205 compound is also confirmed by high values of such indicators as $T_{1/2}$ and MRT, which are 17.7 h and 7.85 h, respectively, as well as a low Cl indicator (1.81 l/h/kg).

To characterize the distribution of the medicinal substance, the index of the apparent volume of distribution (AVD) was calculated. This makes it possible to determine one of the distribution options for the compound: being in the blood plasma without leaving the vascular bed (with the indicator less than the plasma volume); the distribution in the extracellular and intracellular fluid (with comparable indicators with the total amount of fluid in the body) or being mainly in the tissues (with the values higher than the total volume of the body fluid). The obtained value – 43.88 l/kg – exceeds the real volume of fluid in the body of rats (0.67 l/kg) by more than 65 times, which may indicate intensive penetration of the compound into the organs and tissues of the body and deposition in peripheral tissues [17].

The absolute bioavailability was determined by a comparative study of the concentrations dynamics of the test substance in the blood plasma after oral and intravenous administrations. This indicator was calculated as the ratio of the area under the curve for the oral administration of the drug substance to the area under the curve for the intravenous route. Based on the previous studies, the AUC index for the intravenous administration of RU-1205 is 14.76 μg*hour/ml [18], based on which the absolute bioavailability in the oral route corresponds to 37.3%.

The main result of the distribution processes of the drug in the body is its further transport to the zone of a potential action, where it interacts with specific targets, which are both the whole organ and individual cells or specific molecular structures that determine the pharmacological effect of the drug. The penetration intensity of the studied substance into peripheral tissues is characterized by tissue accessibility. The RU-1205 compound is intensively distributed in the tissues of the studied organs, herewith, it was determined that there is a significant heterogeneity in the distribution of the drug among the organs. The analysis of the absolute values of tissue availability (fT) of RU-1205 showed that it has the least tropism to the heart, lungs, spleen, and muscles. In the liver and kidneys, there is a significant content of the studied substance, with AUC values of 17.65 and 82.94 μg*hour/ml, respectively. The tissue distribution parameter for renal tissue was 3.01 and 0.64 for liver tissue, respectively. The substance under study is determined in these target organs for 12 hours in the liver and 8 hours in the kidneys. The highest content was noted in the kidney tissues, which may indicate the predominant renal excretion of RU-1205. In high concentrations, the compound is also determined in the adipose tissue, in the omentum, which, apparently, is due to its lipophilicity. In the brain, after the oral administration, the content of the studied substance is below the detection threshold.

In the study of the RU-1205 excretion after the oral administration, it was determined that the excretion of the compound occurs via renal excretion and through the intestine. Therefore, the test compound is excreted in the urine for four days and in the feces for two days. The intensity of the excretion process during the next three days of the study after the oral administration of the compound is presented by the data of cumulative urinary and intestinal kinds of excretion (Fig. 4). The cumulative urinary excretion is 65.12 μg, which corresponds to about 0.65% of the administered dose. It was also found that more of the studied substance is detected in urine than in feces (Fig. 4 A.). At the same time, it was revealed that the extrarenal (metabolic) clearance of the RU-1205 compound significantly predominates over the renal, which, according to the literature data, indicates an active metabolism of the studied substance in the liver [19, 20].

The study of the analgesic properties of the compound showed that a significant analgesic effect was observed already 15 minutes after the oral administration of RU-1205. In this case, the latent period of the nociceptive reaction increases by 16%. The antinociceptive effect gradually increases and reaches its highest values by the 4th hour after the administration, then gradually decreases, reaching about 50% of the maximum efficiency indicators by 8 hours of observation (Fig. 5). The observed analgesic effect is long lasting, keeping for 12 hours.

A computer projection of the studied substance metabolites *in silico* using the PALLAS 3.00 program, made it possible to determine seven possible metabolites. These metabolites are mainly products of oxidation reactions,

in particular, 3 predicted metabolites under the codes "a", "d" and "g", possibly formed as results of the hydroxylation reaction (Table 1).

DISCUSSION

In recent years, one of the promising groups of opioid analgesics with a selective mechanism of action without any risk of developing respiratory depression and drug dependence (characteristic of μ-agonists), has been considered selective kappa-opioid agonists. In contrast to μ- or δ-agonists, along with a high analgesic activity, they do not stimulate the dopaminergic "reward" system [6, 21-23]. Although kappa-opioid receptor agonists have been long recognized as analgesics with a low abuse potential, serious side effects associated with dysphoria, anhedonia, and hallucinations are a limiting factor in the promotion of first-generation kappa-selective analgesics [24].

Nowadays, there are opioid receptor agonists that can exert an antinociceptive effect without causing undesirable psychotropic effects. The possibility of such an activity is explained by a selective activation of signal transduction pathways from the opioid receptor (selective functional signaling). For example, the stimulation of μ-opioid receptors by morphine, and κ-receptors by the selective kappa agonist U 50488, activates two intracellular cascades Gi/0 and β – arrestin [25, 26]. This is realized in the effective pain relief, as well as side effects in the form of euphoria, dependence formation, respiratory depression (for morphine) and dysphoria (for U 50488). Antinociceptive effects upon the activation of any of the opioid receptors, are realized through the activation of the Gi-protein pathway, while the addictive potential, dysphoria, and most other adverse concomitant effects of opioids, are due to the β-arrestin cascade of intracellular signals. Over the past decade, Gi-biased μ- and κ-opioid agonists (PZM21, Oliceridine, Herkinorin), κ-agonists with a β-arrestin-biased antagonistic activity, as well as ligands (Noribogaine) that combine the properties of highly effective Gi- activators and inhibitors of β –arrestin, have been identified [27–29].

The mechanism of dysphoria and aversion formation induced by kappa receptor agonists consists in the activation of mitogen-activated protein kinase p38-MAPK via the β-arrestin signaling pathway [30]. There is a hypothesis that highly selective kappa-receptor agonists, which do not clearly activate or inhibit the p38-MAP-kinase cascade of intracellular signaling reactions, the stimulation of which is realized in kappa-mediated aversion, hyperalgesia and inflammation, will have a more pronounced analgesic effect without any risk of developing dysphoria [31]. This hypothesis served as a prerequisite for the development of a new technology for searching for kappa-selective agonists with the properties of p38-MAPK inhibitors and the prospect of creating competitive analgesics on their basis without respiratory distress syndrome, narcogenic potential and dysphoria. Most of the selective kappa receptor ligands and selective inhibitors of p38-MAPK are derivatives of cyclic nitrogen-containing heterosystems, which also include benzimidazole derivatives and condensed systems based on it [32].

As a result of a targeted complex study of benzimidazole derivatives using *in silico* computer modeling methods, as well as in the course of experimental studies *in vitro* and *in vivo*, the main regularities of kappa-receptor interactions were determined. They were implemented in an integral scaffold (2-p-fluorophenylimidazo [1, 2-a] benzimidazole) [33–35] and made it possible to identify the original molecule – a compound under the laboratory code – RU-1205. The kappa-receptor mechanism of the compound action was laboratory confirmed in an experimental model of the rabbit vas deferens (the inhibition rate of electrically-induced contraction of the isolated duct IC50 = 2 nM), and was also proven using a non-selective opioid antagonist, naloxone and a kappa-selective antagonist, norbinoltorofimine, which blocked the analgesic activity of the test compound in *in vivo* tests [36].

When administered orally at the dose of 50 mg/kg, the maximum concentration of the RU-1205 compound was observed 1 hour after the administration – the time required for penetration through the gastrointestinal tract wall and through the hepatic barrier. The substance under study circulates for a long time in the blood plasma for 12 hours. The long-term nature of the RU-1205 compound elimination, is also confirmed by high values of its half-life and the average hold-time of the RU-1205 molecule in the body. It is noted that the absolute bioavailability of the substance of the RU-1205 compound when administered orally, is 37%. For comparison, the kappa-opioid receptor agonist butorphanol tartrate used in the animal experiment clinic, has a low bioavailability value after the oral administration (it is less than 10%).

The obtained results of the excretion study showed that the extrarenal (metabolic) clearance of the RU-1205 compound significantly prevails over the renal one. This is consistent with the literature data on the excretion of benzimidazole derivatives. Consequently, benzimidazole derivatives undergo intensive metabolic transformations in the body. It has been shown that afobazole (fabomotizole), after the administration by various methods, is registered in urine and feces only in insignificant amounts [37, 38], 80% of the administered dose of omeprazole is excreted in the urine as metabolites and a small part in feces [39].

When evaluating the computer projection of possible RU-1205 metabolites, it should be noted that in all the compounds formed by the radical, the C^2 atom retains the fluorophenyl radical, which is presumably in-

DOI: 10.19163/2307-9266-2021-9-2-149-160

volved in the development of the analgesic effect [33]. All metabolites, except metabolites "E" and "F", are characterized by the preservation of the morpholine radical, which is also involved in the development of pain relief.

On the basis of the computer analysis, it was also found out that in the process of the of RU-1205 metabolism, the detachment of the morpholine radical and, probably, a change in the analgesic activity (metabolite "E"), is possible.

Both in preclinical studies and in the further clinical use of new drugs, it is advisable to search for relationships between pharmacokinetic parameters and drug effects. The discussion of such correlations is important for understanding the system of relationships between pharmacokinetic and pharmacodynamic mechanisms in the action of the future drug. Therefore, the next stage of the research was to study the dependence of pharmacodynamic properties (pain relief) on the pharmacokinetics of the RU-1205 compound.

The previous studies have shown that the compound has a dose-dependent analgesic effect, and the average effective dose of RU-1205 for the oral route of administration is 5 mg/kg [8]. The main parameter characterizing the degree of bioavailability of the drug, the area under the pharmacokinetic curve RU-1205, increases linearly with increasing the dose [40]. Taking into account the sensitivity of the developed HPLC method and the possibility of comparing pharmacokinetic and pharmacodynamic data with linear kinetics, the correlation of the analgesic effects and pharmacokinetics at different doses, was assessed. When comparing the pharmacokinetic and analgesic properties, it was determined that the difference in the maximum analgesic effect and the peak concentration of the substance in the plasma is 3 hours. This difference can be explained by the peculiarities of the compound RU-1205 penetration through the blood-brain and hepatic barriers. With this route of administration, RU-1205 enters the brain at the concentrations below the threshold and is not detected when studying the tissue accessibility. However, a central analgesic effect is observed, and it suggests that biotransformation of RU-1205 may result in the formation of active metabolites possessing analgesic properties. The literature data show that some opioid analgesics are characterized by the formation of active metabolites [41]. For example, as a result of glucuronization of morphine, the main metabolites of morphine are formed. One of them, morphine-6-glucuronide, has pronounced analgesic properties that are superior to morphine itself [42]. Another opioid analgesic (of the mixed action), tramadol, is metabolized by N- and O-demethylation, followed by conjugation with glucuronic acid to form an active metabolite, mono-O-desmethyltramadol, which causes a more pronounced activation of mu-receptors in comparison with tramadol itself [41]. Hydromorphone is a semi-synthetic derivative of morphine, its pharmacological properties are similar to morphine, including its biotransformation. It is also extensively metabolized by glucuronization up to hydromorphone-3-glucuronide; the other minor metabolites include unconjugated and conjugated dihydromorphine and dihydroisomorphine, hydromorphone-3-sulfate, norohydromorphone, and nordihydroisomorphine [41]. Its metabolites, dihydromorphine and norohydromorphone, have the same potency as morphine. Methadone is an opioid used in a number of countries as an analgesic, as well as in the treatment of drug addiction (drug trafficking in Russia is prohibited, according to List I of the RF Government Decree No. 681 dated June 30, 1998 (as amended on 12/03/2020)), is a racemic mixture of R- and S-methadone. Moreover, R-methadone has almost 50 times more powerful analgesic effect than S-methadone. As a result of the oxidative biotransformation, it undergoes stereoselective metabolism (N-demethylation), and with the participation of CYP2C19, metabolites α-(3S6S)-methadol and α-(3S6S)-N-desmethylmethadol are formed from the enantiomer of S-methadone, the latter of which has an analgesic activity, comparable to R-methadone (active enantiomer) [42]. The analgesic oxycodone (in the oral form, until recently considered as an alternative to morphine, due to a less pronounced addictive effect) is metabolized with N-demethylation up to noroxycodone and codeine, O-demethylation up to oxymorphone through CYP3A4 and CYP2D6, respectively [41, 42]. In the 3rd position, Oxymorphone is further metabolized by glucuronidation to oxymorphone-3-glucuronide. Oxycodone and noroxycodone itself have a lower affinity, while oxymorphone has a higher affinity to the μ-opioid receptor [43, 44].

CONCLUSION

Thus, as a result of the research, the main pharmacokinetic parameters of RU-1205 have been studied at a single oral administration. The processes of the tissue distribution of the studied compound in the organs of the rat organism, as well as its excretion, have been investigated. The value of its absolute bioavailability for this route of administration was 37.3%. Despite the fact that under the developed chromatographic conditions, the RU-1205 compound has not been detected in the brain in the oral route administration, the observed central analgesic effect (realized as a result of penetration through the blood-brain barrier) may indicate a possible presence of active metabolic products with the analgesic activity. The totality of the results obtained suggests that the RU-1205 compound undergoes active biotransformation processes in the body of animals. Based on the revealed pharmacokinetic parameters of the test compound, it is possible to plan an optimal scheme for further clinical trials.

FUNDING

The synthesis of the RU-1205 compound was carried out with the financial support of the Ministry of Science and Higher Education of the Russian Federation within the framework of the State Assignment, grant FENW-2020-0031 (0852-2020-0031)

CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHORS CONTRIBUTION

Alexander A. Spasov – concept and design of the study, approval of the final version of the article; Lyudmila A. Smirnova – development of the experiment, text editing; Olesya Yu. Grechko – administrative organization of the experimen; Natalya V. Eliseeva – analysis of factual material, text writing; Yulia V. Lifanova – sample preparation, statistical processing; Andrey I. Rashchenko – conducting the experimental series to study pharmacokinetic parameters, results processing; Anatoly S. Morkovnik – synthesis of the compound, text editing; Olga N. Zhukovskaya, Vera A. Anisimova – synthesis of the compound. All authors participated in the discussion of the results.

REFERENCES

1. Harbaugh CM, Suwanabol PA. Optimizing Pain Control During the Opioid Epidemic. Surg Clin North Am. 2019 Oct;99(5):867–883. DOI: 10.1016/j.suc.2019.06.002.
2. Arbuck DM, Abuzarova GR, Alekseeva GS. Opioids in pain syndrome management (part 1). Messenger of Anesthesiology and Resuscitation. 2017;14(3):58–67. DOI: 10.21292/2078-5658-2017-14-3-58-67. Russian
3. Wang S. Historical Review: Opiate Addiction and Opioid Receptors. Cell Transplant. 2019 Mar;28(3):233–238. DOI: 10.1177/0963689718811060.
4. Palekhov AV, Vvedenskaya ES. The problems of opioid analgesics provision. The subservice part of the iceberg. Kachestvennaya Klinicheskaya Praktika [Good Clinical Practice]. 2017;(2):43–48. Russian
5. FDA (Food and Drug Administrtion). Analysis of Long-Term Trends in Prescription Opioid Analgesic Products: Quantity, Sales, and Price Trends.FDA Report. 2018: 1–12.
6. Prisinzano TE, Tidgewell K, Harding WW. Kappa opioids as potential treatments for stimulant dependence. AAPS J. 2005 Oct 19;7(3):E592-9. DOI: 10.1208/aapsj070361.
7. Grechko OY, Litvinov RA, Spasov AA, Rashchenko AI, Shtareva DM, Anisimova VA, Minkin VI. Study of μ- and δ-Opioid Activities in Agents with Various κ-Receptor Selectivity. Bull Exp Biol Med. 2017 Mar;162(5):632–635. DOI: 10.1007/s10517-017-3674-5.
8. Spasov AA, Grechko OYu, Shtareva DM, Rashchenko AI, Eliseeva NV, Anisimova VA. Analgesic activity of the kappa opioid receptor agonist – RU-1205 in rats. Journal of Clinical and Health Sciences. 2018; 3(2): 13–18. DOI: 10.24191/jchs.v3i2.7275.
9. Spasov AA, Grechko OYu, Eliseeva NV, Litvinov RA, Shamshina DD. Toxic effect of single treatment with kappa-opioid agonist, RU-1205 compound, on the neurological status of wild type mice. JSM Clinical Pharmaceutics. 2017; 3(1):1014.
10. Thigpen JC, Odle BL, Harirforoosh S. Opioids: A Review of Pharmacokinetics and Pharmacodynamics in Neonates, Infants, and Children. Eur J Drug Metab Pharmacokinet. 2019 Oct;44(5):591–609. DOI: 10.1007/s13318-019-00552-0.
11. Normative legal acts in the field of circulation of medicines in the Eurasian Economic Community. Volume 2. Development and research of medicinal products. M.: Remedium. 2017: 308 p. Russian
12. Su M, Dong C, Wan J, Zhou M. Pharmacokinetics, tissue distribution and excretion study of trans-resveratrol-3-O-glucoside and its two metabolites in rats. Phytomedicine. 2019 May;58:152882. DOI: 10.1016/j.phymed.2019.152882.
13. Smirnova LA, Spasov AA, Raschenko AI, Suchkov EA, Riabuha AF, Kuznetsov KA. Analytical features of chromatographic method of quantitative determination of benzimidazole derivatives. Volgograd Journal of Medical Research. 2013; 2 (38):9–13. Russian.
14. Piotrovsky V.K. Method of statistical moments and integral model-independent parameters of pharmacokinetics. Pharmacology and toxicology [Farmakologiya i toksikologiya]. 1986; 49(5):118–127. Russian
15. Romita VV, Henry JL. Intense peripheral electrical stimulation differentially inhibits tail vs. limb withdrawal reflexes in the rat. Brain Res. 1996 May 13;720(1–2):45–53. DOI: 10.1016/0006-8993(96)00105-9.
16. Millan MJ. Kappa-opioid receptor-mediated antinociception in the rat. I. Comparative actions of mu- and kappa-opioids against noxious thermal, pressure and electrical stimuli. J Pharmacol Exp Ther. 1989 Oct;251(1):334–41.
17. Smith DA, Beaumont K, Maurer TS, Di L. Volume of Distribution in Drug Design. J Med Chem. 2015 Aug 13;58(15):5691–8. DOI: 10.1021/acs.jmedchem.5b00201.
18. Davies B, Morris T. Physiological parameters in laboratory animals and humans. Pharm Res. 1993 Jul;10(7):1093–5. DOI: 10.1023/a:1018943613122.
19. Spasov AA, Smirnova LA, Grechko OU, Raschenko AI, Shtareva DM, Anisimova VA. Pharmacokinetic and analgesic properties of the injectable dosage form of a new imidazobenzimidazole derivative RU-1205 with kappa agonist activity. Biomeditsinskaya khimiya [Biomedical chemistry]. 2015, 61(5), 636–639. Russian
20. Fagerholm U. Prediction of human pharmacokinetics – renal metabolic and excretion clearance. J Pharm Pharmacol. 2007 Nov;59(11):1463–71. DOI: 10.1211/jpp.59.11.0002.

DOI: 10.19163/2307-9266-2021-9-2-149-160

21. Stowe CM, Plaa GL. Extrarenal excretion of drugs and chemicals. Annu Rev Pharmacol. 1968;8:337–56. DOI: 10.1146/annurev.pa.08.040168.002005.
22. Minervini V, Lu HY, Padarti J, Osteicoechea DC, France CP. Interactions between kappa and mu opioid receptor agonists: effects of the ratio of drugs in mixtures. Psychopharmacology (Berl). 2018;235(8):2245–2256. DOI: 10.1007/s00213-018-4920-x.
23. Carlezon WA Jr, Miczek KA. Ascent of the kappa-opioid receptor in psychopharmacology. Psychopharmacology (Berl). 2010 Jun;210(2):107–8. DOI: 10.1007/s00213-010-1849-0.
24. Shippenberg TS, Zapata A, Chefer VI. Dynorphin and the pathophysiology of drug addiction. Pharmacol Ther. 2007 Nov;116(2):306–21. DOI: 10.1016/j.pharmthera.2007.06.011.
25. Margolis EB, Karkhanis AN. Dopaminergic cellular and circuit contributions to kappa opioid receptor mediated aversion. Neurochem Int. 2019 Oct;129:104504. DOI: 10.1016/j.neuint.2019.104504.
26. Siuda ER, Carr R 3rd, Rominger DH, Violin JD. Biased mu-opioid receptor ligands: a promising new generation of pain therapeutics. Curr Opin Pharmacol. 2017 Feb;32:77–84. DOI: 10.1016/j.coph.2016.11.007.
27. Bruchas MR, Chavkin C. Kinase cascades and ligand-directed signaling at the kappa opioid receptor. Psychopharmacology (Berl). 2010 Jun;210(2):137–47. DOI: 10.1007/s00213-010-1806-y.
28. Bedini A, Di Cesare Mannelli L, Micheli L, Baiula M, Vaca G, De Marco R, Gentilucci L, Ghelardini C, Spampinato S. Functional Selectivity and Antinociceptive Effects of a Novel KOPr Agonist. Front Pharmacol. 2020 Mar 5;11:188. DOI: 10.3389/fphar.2020.00188.
29. Maillet EL, Milon N, Heghinian MD, Fishback J, Schürer SC, Garamszegi N, Mash DC. Noribogaine is a G-protein biased κ-opioid receptor agonist. Neuropharmacology. 2015 Dec;99:675–88. DOI: 10.1016/j.neuropharm.2015.08.032.
30. Markham A. Oliceridine: First Approval. Drugs. 2020 Nov;80(16):1739–1744. DOI: 10.1007/s40265-020-01414-9. e. 537, 185–190 (2016). DOI: 10.1038/nature19112.
31. Ehrich JM, Messinger DI, Knakal CR, Kuhar JR, Schattauer SS, Bruchas MR, Zweifel LS, Kieffer BL, Phillips PE, Chavkin C. Kappa Opioid Receptor-Induced Aversion Requires p38 MAPK Activation in VTA Dopamine Neurons. J Neurosci. 2015 Sep 16;35(37):12917–31. DOI: 10.1523/JNEUROSCI.2444-15.2015.
32. Bruchas MR, Land BB, Aita M, Xu M, Barot SK, Li S, Chavkin C. Stress-induced p38 mitogen-activated protein kinase activation mediates kappa-opioid-dependent dysphoria. J Neurosci. 2007 Oct 24;27(43):11614–23. DOI: 10.1523/JNEUROSCI.3769-07.2007.
33. Shanware NP, Williams LM, Bowler MJ, Tibbetts RS. Non-specific in vivo inhibition of CK1 by the pyridinyl imidazole p38 inhibitors SB 203580 and SB 202190. BMB Rep. 2009 Mar 31;42(3):142–7. DOI: 10.5483/bmbrep.2009.42.3.142.
34. Grechko OYu, Spasov AA, Anisimova VA, Petrov VI, Vasiliev PM, Eliseeva NV, Zhukovskaya ON. 2-p-fluorophenylimidazo [1,2-a] benzimidazole is a promising scaffold for the creation of selective kappa opioid agonists. XX Mendeleev Congress on General and Applied chemistry. Yekaterinburg, September 26–30. 2016: 482p. Russian.
35. Yakovlev DS, Stukovina AYu, Goryagin I.I., Grechko O.Yu, Kirillova NV, Kosolapov VA, Tibir'kova EV, Salaznikova OA, Naumenko LV, Gurova NA. Synthesis and pharmacological activity of 9-R-2-halogenophenylimidazo[1,2-a]benzimidazoles. Chemical and Pharmaceutical journal [Khimiko-Farmatsevticheskii Zhurnal]. 2010;44(7):7–13. DOI: 10.30906/0023-1134-2010-44-7-7-13. Russian
36. Spasov AA, Grechko OYu, Eliseeva NV, Vasiliev PM, Anisimova VA. A new class of kappa opioid receptor agonists. Eksperimental'naya i klinicheskaya farmakologiya. 2010; 73(S):8–9. Russian.
37. Seredenin SB, Viglinskaya AO, Kolyvanov GB, Litvin AA, Kravcova OYu, Zherdev VP. Pharmacokinetics of afobazole in rats. Experimental and Clinical Pharmacology [Eksperimental'naya i klinicheskaya farmakologiya]. 2007;70(2):59–64. Russian
38. Seredenin SB, Viglinskaya AO, Mozhaeva TYa, Kolyvanov GB, Litvin AA, Avdyunina NI, Savel'ev VL, Zherdev VP. Afobazole metabolism in rats. Experimental and Clinical Pharmacology [Eksperimental'naya i klinicheskaya farmakologiya]. 2008; 7(2):50–52. DOI: 10.30906/0869-2092-2008-71-2-50-52. Russian
39. Serebrova SYu, Starodubcev AK, Pisarev VV, Kondratenko SN, Vasilenko GF, Dobrovol'skij OV. Pharmacokinetics, duration of the antisecretory effect of omeprazole and esomeprazole, probable causes of their changes in duodenal ulcer. Experimental and Clinical Gastroenterology [Eksperimental'naya i klinicheskaya gastroenterologiya]. 2009;4:86–92. Russian
40. Smirnova LA, Rashchenko AI, Ryabuha AF, Kuznecov KA, Suchkov EA. Quantification of a new agonist for κ-opioid receptors, a benzimidazole derivative [Kolichestvennoe opredelenie novogo agonista k-opioidnyh receptorov proizvodnogo benzimidazola]. Materials of the IV Congress of Russian Pharmacologists "Innovations in modern pharmacology", Kazan, September 18-21. 2012: 171–172. Russian
41. Smith HS. The metabolism of opioid agents and the clinical impact of their active metabolites. Clin J Pain. 2011 Nov-Dec;27(9):824–38. DOI: 10.1097/AJP.0b013e31821d8ac1.
42. Sverrisdóttir E, Lund TM, Olesen AE, Drewes AM, Christrup LL, Kreilgaard M. A review of morphine and morphine-6-glucuronide's pharmacokinetic-pharmacodynamic relationships in experimental and clinical pain. Eur J Pharm Sci. 2015 Jul 10;74:45–62. DOI: 10.1016/j.ejps.2015.03.020.
43. Coller JK, Christrup LL, Somogyi AA. Role of active metabolites in the use of opioids. Eur J Clin Pharmacol. 2009 Feb;65(2):121–39. DOI: 10.1007/s00228-008-0570-y.
44. Lalovic B, Phillips B, Risler LL, Howald W, Shen DD. Quantitative contribution of CYP2D6 and CYP3A to oxycodone metabolism in human liver and intestinal microsomes. Drug Metab Dispos. 2004 Apr;32(4):447–54. DOI: 10.1124/dmd.32.4.447.

ISSN 2307-9266 e-ISSN 2413-2241

Scientific and Practical Journal
PHARMACY & PHARMACOLOGY

AUTHORS

Alexander A. Spasov – Academician of the Russian Academy of Sciences, Doctor of Sciences (Medicine), Professor, Head of the Department of Pharmacology and Bioinformatics of Volgograd State Medical University. ORCID ID: 0000-0002-7185-4826. E-mail: aspasov@mail.ru

Lyudmila A. Smirnova – Doctor of Sciences (Biology), Head of the Laboratory of Pharmacological Kinetics, Research Institute of Pharmacology, Volgograd State Medical University. ORCID ID: 0000-0002-0950-8519. E-mail: smirlusi@yandex.ru

Olesya Yu. Grechko – Doctor of Sciences (Medicine), Professor of the Department of Pharmacology and Bioinformatics of Volgograd State Medical University. ORCID ID: 0000-0002-4184-4897. E-mail: olesiagrechko@mail.ru

Natalya V. Eliseeva – Candidate of Sciences (Medicine), Associate Professor of the Department of Pharmacology and Bioinformatics, Volgograd State Medical University. ORCID ID: 0000-0002-2243-5326. E-mail: nvkirillova@rambler.ru

Yulia V. Lifanova – Assistant of the Department of Pharmacology and Bioinformatics of Volgograd State Medical University. ORCID ID: 0000-0001-9663-5067. E-mail: j_semenova_pharm@mail.ru

Andrey I. Rashchenko – Candidate of Sciences (Pharmacy), Senior Researcher, Laboratory of Pharmacology of Antioxidant Agents, Research Institute of Pharmacology, Volgograd State Medical University. ORCID ID: 0000-0001-8357-4007. E-mail: a.rashencko@yandex.ru

Olga N. Zhukovskaya – Candidate of Sciences (Chemistry), Researcher, Laboratory of Organic Synthesis, Scientific Research Institute of Physical Chemistry, Southern Federal University. ORCID ID: 0000-0003-0865-6656. E-mail: zhukowskaia.ol@yandex.ru

Anatoly S. Morkovnik – Doctor of Sciences (Chemistry), Chief Researcher of the Laboratory of Organic Synthesis, Research Institute of Physical Chemistry, Southern Federal University. ORCID ID: 0000-0002-9182-6101. E-mail: asmorkovnik@sfedu.ru

Vera A. Anisimova – Candidate of Sciences (Chemistry), Senior Researcher of the Laboratory of Organic Research Institute of Physical Chemistry, Southern Federal University. ORCID ID: 0000-0002-0877-4575