DOI: 10.6060/ivkkt.20246702.6901

УДК: 547.876

СИНТЕЗ И АНТИОКИСЛИТЕЛЬНАЯ АКТИВНОСТЬ ПРОИЗВОДНЫХ ТИАДИАЗИНОВ С ФЕНОЛЬНЫМИ ФРАГМЕНТАМИ

В.Н. Кошелев, О.В. Примерова, А.П. Герман, А.А. Гладких

Владимир Николаевич Кошелев (ORCID 0000-0001-5755-5291), Ольга Вячеславовна Примерова (ORCID 0000-0002-0162-2528)*, Алиса Павловна Герман, Анастасия Александровна Гладких (ORCID 0009-0006-3781-0666)

Кафедра органической химии и химии нефти, РГУ нефти и газа (НИУ) им. И.М. Губкина, Ленинский пр., 65, Москва, Российская Федерация, 119991
E-mail: koshelev.v@gubkin.ru, Primerova92@yandex.ru *, anatanielle@yandex.ru, german_al5@mail.ru

Синтезирован ряд тиокарбогидразонов на основе 4-гидрокси-3,5-ди-трет-бутилбензальдегида, салицилового альдегида, ванилина и 2,3-дигидрокси-4,6-ди-трет-бутилбензалдегида. Реакцией между тиокарбогидразонами и бромацетилкумарином получен и охарактеризован ряд производных тиадиазина, содержащих фенольный фрагмент, выходы составили 56-90%. Структурные свойства синтезированных веществ исследовали методами ИК-Фурье, ^{1}H, ^{13}C ЯМР спектроскопии: в ИК-спектрах тиогидразинов присутствуют полосы, характерные для валентных колебаний ОН-группы (3622 см$^{-1}$), N-H связи (3450 см$^{-1}$), двойных связей C=C и C=N 1608 и 1541 см$^{-1}$ соответственно. В отличие от спектра исходных соединений наблюдается пик, соответствующий колебаниям связи C=O (1722 см$^{-1}$), который появился вследствие введения кумаринового фрагмента. В спектрах ^{1}H ЯМР полученных соединений присутствуют пики в области 3,9 м.д., отвечающие протонам тиадиазинового фрагмента, отсутствуют сигналы в области 6,8 м.д., относящиеся к CH протону тиазольного кольца, также присутствует набор пиков в области 7,5-8,4 м.д., характерный для кумаринового кольца. Антиоксидантную активность всех продуктов определяли in vitro: активность по ингибированию катион-радикалов оценивали с использованием 2,2'-азино-бис(3-этилбензотиазолин-6-сульфоната), а электронодонорную активность определяли по способности восстанавливать железо с использованием метода феррицианида/берлинской лазури, в обоих случаях в качестве стандарта был использован агидол. Все испытанные вещества проявили более высокую активность в обоих испытаниях по сравнению со стандартом. Лучшие антиокислительные свойства в обоих методах проявило производное 2,3-дигидрокси-4,6-ди-трет-бутилбензальдегида, высокую антирадикальную активность также показал тиадиазин на основе салицилового альдегида, а хорошие железовосстанавливающие свойства проявил тиадиазин с фрагментом 2-метоксифенола.

Ключевые слова: органический синтез, фенолы, тиадиазины, антиокислители

SYNTHESIS AND ANTIOXIDANT ACTIVITY OF THIADIAZINE DERIVATIVES WITH PHENOLIC FRAGMENTS

V.N. Koshelev, O.V. Primerova, A.P. German, A.A. Gladkikh

Vladimir N. Koshelev (ORCID 0000-0001-5755-5291), Olga V. Primerova (ORCID 0000-0002-0162-2528)*, Alisa P. German, Anastasia A. Gladkikh (ORCID 0009-0006-3781-0666)

Department of Organic Chemistry and Petroleum Chemistry, National University of Oil and Gas «Gubkin University», Leninsky ave., 65, Moscow, 119991, Russia
E-mail: koshelev.v@gubkin.ru, Primerova92@yandex.ru *, anatanielle@yandex.ru, german_al5@mail.ru

A series of thiocarbohydrazones based on 3,5-di-tert-butyl-4-hydroxybenzaldehyde, salicylaldehyde, vanillin and 4,6-di-tert-butyl-2,3-dihydroxybenzaldehyde were synthesized. The reaction between thiocarbohydrazones and bromoacetylcoumarin was used to obtain and characterize

a number of thiadiazine derivatives containing a phenolic fragment. The yields were 56–90%. The structural properties of the synthesized substances were studied by IR-Fourier, ^{1}H, ^{13}C NMR spectroscopy. In the IR spectra of thiadiazines there are characteristic bands of the stretching vibrations of the OH group (3622 cm^{-1}), N-H bond (3450 cm^{-1}), double bonds C=C and C=N 1608 and 1541 cm^{-1}, respectively. In contrast to the spectrum of the starting compounds, there is a peak corresponding to vibrations of the C=O bond (1722 cm^{-1}), which appeared due to the coumarin fragment. The ^{1}H NMR spectra of the obtained compounds contain peaks at 3.9 ppm, corresponding to the protons of the thiadiazine fragment. There are no signals at 6.8 ppm, related to the CH proton of the thiazole ring, and there is also a set of peaks in the region of 7.5-8.4 ppm., characteristic of the coumarin ring. Antioxidant activity of all products was determined in vitro: radical cation inhibition activity was determined using 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonate) and electron donating activity was determined by ferric reduction ability using the ferricyanide/Prussian blue method. In both cases, agidol was used as a standard. All tested substances showed higher activity in both tests compared to the standard. The best antioxidant properties in both methods were shown by the derivative of 4,6-di-tert-butyl-2,3-dihydroxybenzaldehyde, thiadiazine based on salicylaldehyde also showed high antiradical activity, and thiadiazine with a fragment of 2-methoxyphenol showed good ferric-reducing properties.

Key words: organic synthesis, phenols, thiadiazines, antioxidants

Для цитирования:
Кошелев В.Н., Примерова О.В., Герман А.П., Гладких А.А. Синтез и антиокислительная активность производных тиадиазинов с фенольными фрагментами. *Изв. вузов. Химия и хим. технология*. 2024. Т. 67. Вып. 2. С. 74–80. DOI: 10.6060/ivkkt.20246702.6901.

For citation:
Koshelev V.N., Primerova O.V., German A.P., Gladkikh A.A. Synthesis and antioxidant activity of thiadiazine derivatives with phenolic fragments. *ChemChemTech* [*Izv. Vyssh. Uchebn. Zaved. Khim. Khim. Tekhnol*.]. 2024. V. 67. N 2. P. 74–80. DOI: 10.6060/ivkkt.20246702.6901.

INTRODUCTION

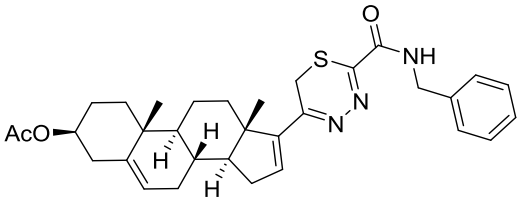
Reactive oxygen species (ROS) are reactive oxygen-containing molecules formed as a result of cellular metabolism. They play an important role in homeostasis, cellular signaling, but they also cause oxidative damage under certain stressful environmental conditions, such as exposure to heavy metals, UV radiation, while the balance necessary for normal cellular homeostasis is disturbed [1]. The overproduction of reactive oxygen species is associated with the development of various degenerative diseases such as cancer [2-5], as well as respiratory, neurodegenerative and digestive diseases [6, 7]. Substances that delay or prevent substrate oxidation are called antioxidants. A lot of research is currently underway to develop substances that exhibit powerful antioxidant activity [8-10].

In this regard, thiocarbohydrazones and their heterocyclic derivatives are of great interest. Thiocarbohydrazone derivatives are used to create antioxidants that suppress reactive oxygen species that cause physiological disorders in the human body [11-13]. It has been shown that derivatives obtained by condensation of thiocarbohydrazide with 4-hydroxy-3-ethoxybenzaldehyde and 4-hydroxy-3,5-dimethoxybenzaldehyde have the highest activity [12]. In addition, thiocarbohydrazide derivatives exhibit high activity against gram-positive and gram-negative bacteria, fungal strains, bacteria of the Mycobacterium tuberculosis complex group [14-18]. Dithiocarbohydrazones derived from salicylaldehyde and vanillin showed good antibacterial activity against gram-negative bacterium Escherichia coli. It was reported that complexes of thiocarbohydrazones with various metal ions have antibacterial and antifungal effects, and can also be used in the future to create drugs for the treatment of neurodegenerative diseases [15].

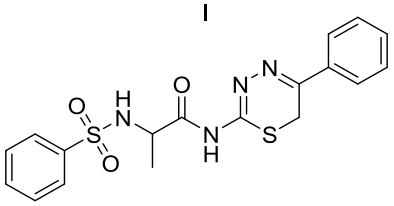
Thiocarbogirazones are widely used in the synthesis of heterocyclic compounds that exhibit antioxidant properties: thiazolinones [19], 1,3-oxadiazolan-5-ones [20], isatin derivatives [21]. Compounds containing thiadiazine fragments have anticancer activity, for example, compound I containing a steroid fragment exhibits high cytotoxicity against prostate cancer cells [22]. Thiadiazine II showed high inhibitory activity against various matrix metalloproteinases (with high affinity for matrix metalloproteinase 9), which are overexpressed in various types of cancer, including lung cancer, and play a crucial role in tumor metastasis [23].

In addition to a wide range of biological activity, thiadiazines also have good antioxidant properties [24, 25], however, the scientific literature contains a

small number of systematic studies on the antioxidant activity of thiadiazines derived from thiocarbohydrazones. The aim of this work is the synthesis and study of the antioxidant properties of thiadiazines based on thiocarbohydrazones with phenolic fragments.



I



II

Fig. 1. Thiadiazines showing high anticancer activity
Рис. 1. Тиадиазины, проявляющие высокую противораковую активность

EXPERIMENTS AND METHODS

Reagents and solvents were purchased from Acros and Sigma-Aldrich. Melting points were determined using a Stuart SMP30 instrument. IR spectra were recorded using an Agilent Carry 600 spectrometer equipped with an attenuated total reflection (ATR) device. ^{1}H and ^{13}C NMR spectra were recorded at room temperature on a Bruker Avance II 300 spectrometer (^{1}H, 300 MHz; ^{13}C, 75 MHz). Me_4Si was used as an internal standard. Elemental analysis was performed on a Vario MicroCube instrument. Antioxidant activity was determined using a UV-Vis spectrophotometer PB-2201.

Synthesis of thiocarbohydrazones 1a-d

2 mmol of aldehyde was dissolved in ethyl alcohol (20 ml), 2.4 mmol of thiocarbohydrazide and three drops of acetic acid were added. The reaction was carried out for 2 h with constant stirring and reflux. After cooling, the resulting precipitate was filtered off.

1a N'-(3,5-di-tert-butyl-4-hydroxybenzylidene)hydrazinecarbothiohydrazide

White crystals, m.p. 201-203 °C (yield 74%). ^{1}H NMR (DMSO-d6, δ, ppm, J_{HH}, Hz): 11.22 s (1H, NH), 9.65 s (1H, NH), 7.93 s (1H, CH), 7.45 s (2H, H-Ar), 7.33 s (1H, OH), 4.84 s (2H, NH_2). FT-IR, ν, cm^{-1}: ν_{OH} – 3600, ν_{C-O} – 1010, ν_{NH} – 3174, 3228, $\nu_{C=C}$ – 1234, $\nu_{C=N}$ – 1514. Calc., %: C, 59.59; H, 8.13; N, 17.37; S, 9.94. Found, %: C, 59.41; H, 8.35; N, 17.32; S, 9.91.

1b N'-(2-hydroxybenzylidene)hydrazinecarbothiohydrazide

White crystals, m.p. 217-218 °C (lit. 217-218 °C [26]) (yield 67%). NMR ^{1}H (DMSO-d6, δ, ppm, J_{HH}, Hz): 11.35 s (1H, NH), 9.87 s (1H, OH), 8.30 s (1H, CH=N), 7.93 d (^{3}J = 9, 1H, H-Ar), 6.70-6.77 m (3H, Har). ^{13}C NMR (DMSO-d6, δ, ppm): 175.8, 156.4, 140.4, 131.8, 127.0, 121.5, 119.1, 116.2. FT-IR, ν, cm^{-1}: ν_{OH} – 3600, ν_{NH} – 3075, $\nu_{C=N}$ – 1615, ν_{C-O} – 1010. Calc., %: C, 45.70; H, 4.79; N, 26.65; S, 15.25. Found, %: C, 45.52; H, 4.88; N, 26.64; S, 15.20.

1c N'-(4- hydroxy-3-methoxybenzylidene)hydrazinecarbothiohydrazide

White crystals, m.p. 197-198 °C (yield 75%). ^{1}H NMR (DMSO-d6, δ, ppm, J_{HH}, Hz): 11.3 s (1H, OH), 9.8 s (1H, NH), 9.4 s (1H, NH), 7.9 s (1H, N=CH), 7.6 s (1H, H-Ar), 7.0 d (^{3}J = 9, 1H, Har), 1.4 s (9H, t-Bu), 6.8 d (^{3}J = 9Hz, 1H, H-Ar), 3.9 s (3H, OCH_3). ^{13}C NMR (DMSO-d6, δ, ppm): 176.4, 149.1, 148.6, 143.3, 126.1, 123.0, 115.6, 109.6, 56.3. FT-IR, ν, cm^{-1}: ν_{OH} – 3420, ν_{C-O} – 1010, ν_{NH} – 3190, 3220, $\nu_{C=N}$ – 1620. Calc., %: C, 44.99; H, 5.03; N, 23.32; S, 13.34 Found, %: C 44.75, H 5.12, N 23.16, S 13.36

1d N'-(4,6-di-tert-butyl-2,3-dihydroxybenzylidene)hydrazinecarbothiohydrazide

White crystals, m.p. 228-230°C (yield 73%). ^{1}H NMR (DMSO-d6, δ, ppm, J_{HH}, Hz): 13.15 s (1H, OH), 9.36 s (2H, NH_2), 8.23 s (1H, N=CH), 6.75 s (1H, H-Ar), 1.39 s (9H, t-Bu), 1.36 s (9H, t-Bu). ^{13}C NMR (DMSO-d_6, *δ*, ppm): 181.7, 172.0, 147.7, 142.7, 138.6, 135.3, 114.2, 113.6, 35.6, 35.2, 32.9, 29.6. FT-IR, ν, cm^{-1}: ν_{OH} – 3600, ν_{C-O} – 1010, ν_{NH} – 3174, 3228, $\nu_{C=C}$ – 1234, $\nu_{C=N}$ – 1514. Calc., %: C 45.49; H 4.29; N 19.89; S, 15.18. Found, %: C 45.62, H 4.43, N 19.76, S 15.03

Synthesis of thiadiazines 2a-d

To 1.5 mmol of thiocarbohydrazone was added 20 ml of isopropyl alcohol and 1.5 mmol of 3-bromoacetylcoumarin. The reaction was carried out for 2 h with constant stirring under reflux. After cooling, the resulting precipitate was filtered off and dried. The compounds were purified by column chromatography on silica gel (DuraSil N with a particle size of 40-60 microns), dichloroethane:ethanol 10:1 mixture was used as an eluent.

2a 3-(2-(2-(4-hydroxy-3,5-di-tert-butyl-4-hydroxybenzylidene)hydrazinyl)-6H-1,3,4-thiadiazin-5-yl)-2H-chromen-2-one

Yellow crystals, m.p. 157-159 °C. (yield 90%). ^{1}H NMR (DMSO-d_6, δ, ppm, J_{HH}, Hz): 9.98 s (1H, CH=N), 8.55 s (1H, H-Ar coumarin), 8.27 d (^{2}J = 4, 1H, H-Ar coumarin), 8.07 t (^{3}J = 6, 1H, H-Ar coumarin), 7.87 d (^{2}J = 4, 1H, H-Ar coumarin), 7.69 s (2H, H-Ar phenol), 7.48 s (1H, OH),) 7.47- 7.34 m (1H, H-

Ar coumarin), 3.89 s (2H, CH_2), 1.42 s (18H, t-Bu). ^{13}C NMR (DMSO-d6, δ, ppm): 192.5, 168.4, 160.4, 158.9, 156.0, 143.4, 140.3, 139.7, 139.5, 139.1, 129.4, 128.7, 127.3, 123.6, 116.4, 110.9, 102.6, 34.9, 30.6, 30.2. FT-IR, ν, cm^{-1}: $\nu_{C=O}$ – 1716 , ν_{C-N} – 1097 [1], $\nu_{C=C}$ – 1664 , $\nu_{C=N}$ – 1577.

2b 3-(2-(2-(2-hydroxybenzylidene)hydrazinyl)-6H-1,3,4-thiadiazin-5-yl)-2H-chromen-2-one

Yellow crystals, m.p. 238-240 °C (yield 56%). ^1H NMR (DMSO-d6, δ, ppm, J_{HH}, Hz): 11.91 s (1H, OH), 8.58 s (1H, CH), 7.90 d (^3J=9, 1H, H-Ar), 7.72 t (^3J = 9,1H, H-Ar), 7.55 - 7.39 m (^3J = 9, 4H, H-Ar), 7.34 t (^3J = 4, 1H, H-Ar), 6.98-6.90 m (^3J = 3, 2H, H-Ar), 3.83 s (2H, CH_2). ^{13}C NMR (DMSO-d6, δ, ppm): 161.6, 159.7, 158.4, 155.9, 153.9, 144.9, 142.1, 133.2, 131.9, 131.2, 129.8, 125.3, 123.9, 120.0, 119.8, 119.3, 119.2, 116.6, 116.5. FT-IR, ν, cm^{-1}: $\nu_{C=O}$ – 1712 , $\nu_{C=C}$ – 1612 , $\nu_{C=N}$ – 1535 . Calc., %: C, 60.31; H, 3.73; N, 14.81; S, 8.47. Found, %: C, 60.12; H, 3.95; N, 14.78; S, 8.42.

2c 3-(2-(2-(4-hydroxy-3-methoxybenzylidene)hydrazinyl)-6H-1,3,4-thiadiazin-5-yl)-2H-chromen-2-one

Yellow crystals, m.p. 177-180 °C (yield 80%). ^1H NMR (DMSO-d6, δ, ppm, J_{HH}, Hz): 9.87 s (1H, OH), 8.54 (1H, CH=N), 8.46 s (1H, H-Ar coumarin), 7.86 d (^3J = 9, 1H, H-Ar coumarin), 7.53- 7.29 m (^3J = 6, 4H, H-Ar), 7.20- 7.11 m (^3J = 6, 1H, H-Ar), 6.90-6.78 m (^3J = 9, 1H, H-Ar), 3.85 s (2H, CH_2), 3.83 s (3H, O-CH_3). ^{13}C NMR (DMSO-d6, δ, ppm): 167.6, 159.3, 154.0, 152.1, 150.6, 148.5, 146.7, 137.2, 133.9, 129.8, 125.6, 125.3, 124.5, 123.0, 118.8, 116.8, 116.2, 110.4, 108.8, 56.0. FT-IR, ν, cm^{-1}: $\nu_{C=O}$ – 1710 , $\nu_{C=C}$ – 1608 , $\nu_{C=N}$ – 1523 , ν_{C-O-C} – 1241 . Calc., %: C, 58.82; H, 3.95; N, 13.72; S, 7.85. Found, %: C, 58.60; H, 3.88; N, 13.66; S, 7.82.

2d 3-(2-(2-(4,6-di-tert-butyl-2,3-dihydroxybenzylidene)hydrazienyl)-6H-1,3,4-thiadiazin-5-yl)-2H-chromen-2-one

Yellow crystals, m.p. 227-229 °C (yield 76%). ^1H NMR (DMSO-d6, δ, ppm, J_{HH}, Hz): 12.65 s (1H, OH), 11.97 s (1H, OH), 9.29 s (1H, N=CH), 8.36 s (1H, H-Ar coumarin), 7.91- 7.88 dd (^2J = 2, ^3J= 8, 1H, H-Ar, coumarin), 7.66 d (^3J = 9, 1H, H-Ar coumarin), 7.53-7.42 m (^3J = 9, 2H, H-Ar, coumarin), 6.83 s (1H, H-Ar), 4.03 s (2H, CH_2), 1.42 s (9H, t-Bu), 1.37 s (9H, t-Bu). ^{13}C NMR (DMSO-d6, δ, ppm): δ 174.8, 159.6, 156.3, 154.0, 149.0, 147.6, 144.8, 142.7, 139.0, 138.7, 136.7, 136.3, 129.8, 125.4, 123.8, 119.3, 116.5, 114.7, 113.3, 35.5, 35.3, 33.1, 29.5. FT-IR, ν, cm^{-1}: ν_{OH} – 3615, $\nu_{C=O}$ – 1730 , $\nu_{C=C}$ – 1617 . Calc., %: C, 64.01; H, 5.97; N, 11.06; S, 6.33. Found, %: C, 64.01; H, 5.97; N, 11.06; S, 6.33.

Antioxidant activity

ABTS assay 2,2'-Azinobis(3-ethylbenzothiazoline-6-sulfonic acid) (Sigma) radical cation ($ABTS^{+\cdot}$) was produced by reacting 7 mM ABTS water solution with 2.45 mM potassium persulfate and allowing the mixture to stand in the dark at room temperature for 12-16 h before use. A dark blue color should be developed. The working solution was prepared by taking a volume of the previous solution and diluting it in ethanol until its absorbance was 0.70 ± 0.02 at 734 nm. To 2.7 ml of working solutions 300 μl of 1 mmol solution of prepared compounds in DMSO were added. After 10 min the absorbance at 734 nm was measured using a spectrofotometer. The percentage inhibition calculated as ABTS radical scavenging activity (%)=(Abscontrol−Abssample)/Abscontrol where Abscontrol is the absorbance of ABTS radical in methanol; Abssample is the absorbance of ABTS radical solution mixed with sample extract/standard. All determinations were performed in triplicate.

Ferric ion-reducing capacity assay. Sample dilutions were prepared in 50 mM phosphate buffer, pH 7.0 and 500 μL of dilutions were mixed with 250 μL of 1% potassium ferricyanide solution followed by incubation for 20 min at 50 °C. After the incubation 500 μL of 10% trichloroacetic acid was mixed with 500 μL of the incubated sample, 100 μL of 0.1% ferric chloride and 500 μL of distilled water. The mixture was left to incubate for 10 min at room temperature and the absorbance was immediately measured at 700 nm, against blank, which consisted of phosphate buffer and appropriate volume of solvent. The results were expressed as absorbance units at 700 nm which was considered as a measure of reducing power.

RESULTS

The synthesis of target compounds was carried out in accordance with Fig. 2, 3. At the first stage, thiocarbohydrazones were obtained by reacting of thiocarbohydrazide with a number of aromatic aldehydes: 4-hydroxy-3,5-di-tert-butylbenzaldehyde, salicylic aldehyde, vanillin, 2,3-dihydroxy-4,6-di-tert-butylbenzaldehyde, and thiocarbohydrazide. The reaction was carried out by reflux the initial reagents in ethanol with the addition of catalytic amounts of acetic acid for 2 h, as described in [27].

The yields of thiocarbohydrazones were 63-74%. The IR-spectrum of the thiocarbohydrazone of 4-hydroxy-3,5-di-tert-butylbenzaldehyde **1d** is typical for compounds **1a-d**. Since the substance contains a phenolic fragment, there is a peak corresponding to the stretching vibrations of the OH group (3600 cm^{-1}) and the stretching vibrations of the C-O bond (1010 cm^{-1}).

It is possible to distinguish peaks of stretching vibrations of bonds in amino groups (3174, 3228 cm-1). Intense peaks are also observed at 1534 cm^{-1} and 1514 cm^{-1}, which are characteristic of C=C and C=N double bonds, respectively [28, 29].

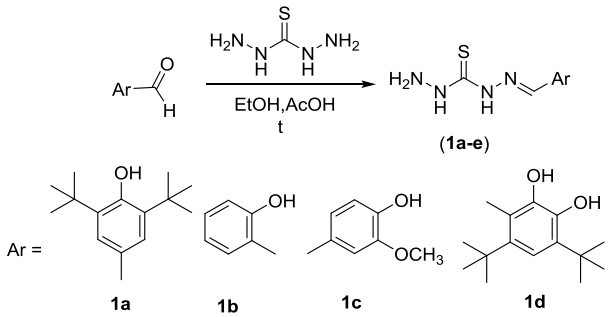


Fig. 2. Synthesis of thiocarbohydrazones **1a-d**
Рис. 2. Схема синтеза тиокарбогидразонов **1a-d**

The reaction of thiocarbohydrazones with halogenoketones can lead to the formation of thiadiazines or aminothiazoles, which is mainly determined by the structure of the starting thiocarbohydrazone [30, 31]. During the cyclization of the thiocarbohydrazones with phenolic fragments with 3-bromoacetylcoumarin in isopropyl alcohol by reflux for 2 h [32, 33] thiadiazines **2a-d** were obtained in 56-82% yields.

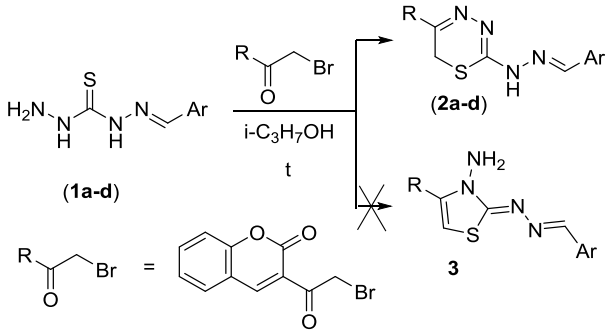


Fig. 3. Synthesis of thiadiazine **2a-d**
Рис. 3. Схема синтеза тиадиазинов **2a-d**

In the IR spectra of thiadiazines, there are bands of the stretching vibrations of the OH group (3622 cm^{-1}), N-H group (3450 cm^{-1}), C=C and C=N double bonds 1608 and 1541 cm^{-1}, respectively. In contrast to the spectrum of the starting compounds **1a-d**, a peak corresponding to the vibrations of the C=O bond is observed (1722 cm^{-1}), which appeared due to the incorporation of the coumarin fragment. The ^1H NMR spectra of the obtained compounds contain peaks at 3.9 ppm, corresponding to the protons of the thiadiazine fragment, there are no signals at 6.8 ppm, related to the CH proton of the thiazole ring, and there is also a set of peaks in the region of 7.5-8.4 ppm., characteristic of the coumarin ring.

The antioxidant activity of the obtained compounds was investigated by the ABTS and PFRAP methods, which are based on electron transfer reactions. The antioxidant activity is estimated by spectrophotometric methods based on the change in the color intensity of the solution during the reduction of oxidizing agents. The change in color depends on the concentration of the antioxidant in the test sample [34].

The ABTS method is based on the formation of the ABTS radical cation. (2,2'-Azino-bis(3-ethylbenzthiazoline-6-sulfonic acid)) due to the loss of an electron by the nitrogen atom during the oxidation of the solution with potassium persulfate. The resulting solution has a bright blue color and absorbs radiation at a wavelength of 743 nm. In the presence of antioxidants, the radical cation is inhibited, and a decrease in their amount is accompanied by discoloration of the solution [35]. The results obtained are shown in Fig. 2.

All synthesized substances showed antiradical activity higher than that of agidol chosen as a standard. At the same time, the best results were observed for thiadiazine obtained on the basis of 2,3-dihydroxy-4,6-di-tert-butylbenzaldehyde **2d**, which is explained by the presence of a larger number of hydroxy groups capable of scavenging radicals.

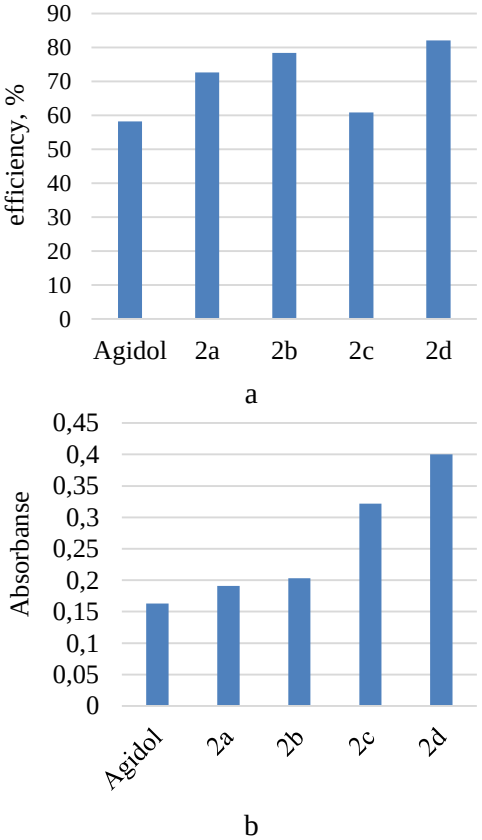


Fig. 4. The results of the study of antioxidant capacity: a) ABTS, б) PFRAP
Рис. 4. Результаты исследования антиокислительной способности: а) метод ABTS, б) метод PFRAP

The study of the reducing ability of thiadiazines was carried out in relation to Fe (III) ions and potassium hexacyanoferrate (III). When antioxidants are added to the working solution, Fe is reduced with the formation of Prussian blue.

It can be seen from the data presented that thiazines obtained from vanillin (**2c**) and 2,3-dihydroxy-4,6-di-tert-butylbenzaldehyde (**2d**) showed the best reducing ability with respect to Fe (III) ions, which can be explained by the presence of strong electron-donating groups, which is associated with a high ionization potential of these molecules, as well as the stability of the radicals formed from them.

CONCLUSIONS

A number of new thiadiazines based on thiocarbohydrazones of salicylaldehyde, vanillin, 2,6-di-tert-butylphenol, and alkylated pyrocatechol have been synthesized. The structural properties of the synthesized substances were studied by IR-Fourier and ^{1}H, ^{13}C NMR spectroscopy. The data obtained showed that thiadiazines derived from thiocarbohydrazones can be effective antioxidants. All synthesized compounds showed high efficiency both in the inhibition of radical cations and in the evaluation of electron-donating properties. The best antioxidant properties in both methods were shown by the derivative of 2,3-dihydroxy-4,6-di-tert-butylbenzaldehyde, thiadiazine based on salicylaldehyde also showed high antiradical activity, and thiadiazine with a fragment of 2-methoxyphenol showed good ferric-reducing properties.

The authors declare the absence a conflict of interest warranting disclosure in this article.

Авторы заявляют об отсутствии конфликта интересов, требующего раскрытия в данной статье.

REFERENCES
ЛИТЕРАТУРА

1. **Das K., Roychoudhury A.** Reactive oxygen species (ROS) and response of antioxidants as ROS-scavengers during environmental stress in plants. *Front. Environ. Sci.* 2014. V. 2. P. 53. DOI: 10.3389/fenvs.2014.00053.
2. **Harris I.S., DeNicola G.M.** The complex interplay between antioxidants and ROS in cancer. *Trends Cell Biol.* 2020. V. 30. N 6. P. 440-451. DOI: 10.3390/antiox11101995.
3. **Kim S.J., Kim H.S., Seo Y.R.** Understanding of ROS-inducing strategy in anticancer therapy. *Oxid. Med. Cell. Longev.* 2019. V. 2019. DOI: 10.1155/2019/5381692.
4. **Белая Н.И., Белый А.В., Клецова В.А.** Хроновольтамперометрическое исследование антирадикальных свойств флавоноидов в реакции с активными формами кислорода. *Изв. вузов. Химия и хим. технология.* 2020. Т. 63. Вып. 9. С. 43-48. DOI: 10.6060/ivkkt.20206309.6217.
   Belaya N.I., Belyi A.V., Kletsova V.A. Chronovoltammetric study of antiradical properties of flavonoids in reaction with reactive oxygen species. *ChemChemTech [Izv. Vyssh. Uchebn. Zaved. Khim. Khim. Tekhnol.]*. 2020. V. 63. N 9. P. 43-48 (in Russian). DOI: 10.6060/ivkkt.20206309.6217.
5. **Сахабутдинова Г.Н., Раскильдина Г.З., Мещерякова С.А., Шумадалова А.В., Борцова Ю.Л., Кузьмина У.Ш., Злотский С.С., Султанова Р.М.** Антиоксидантная и цитотоксическая активность ряда O- и S-содержащих макроциклов. *Изв. вузов. Химия и хим. технология.* 2020. Т. 63. Вып. 3. С. 82-87. DOI: 10.6060/ivkkt.20206303.6118.
   Raskil'dina G.Z., Sakhabutdinova G.N., Meshcheryakova S.A., Shumadalova A.V., Bortsova Yu.L., Kuzmina U.Sh., Zlotsky S.S., Sultanova R.M. Antioxidant and cytotoxic activity of a series of O- and S-containing macrocycles. *ChemChemTech [Izv. Vyssh. Uchebn. Zaved. Khim. Khim. Tekhnol.]*. 2020. V. 63. N 3. P. 82-87 (in Russian). DOI: 10.6060/ivkkt.20206303.6118.
6. **Liu Z., Ren Z., Zhang J., Chuang C.-C., Kandaswamy E., Zhou T., Zuo L.** Role of ROS and nutritional antioxidants in human diseases. *Front. Physiol.* 2018. V. 9. P. 477. DOI: 10.3389/fphys.2018.00477.
7. **Morrell C.N.** Reactive oxygen species: finding the right balance. *Circulation Res.* 2008. V. 103. N 6. P. 571–572. DOI: 10.1161/CIRCRESAHA.108.184325.
8. **Santos-Sánchez N.F., Salas-Coronado R., Villanueva-Cañongo C., Hernández-Carlos B.** Antioxidant compounds and their antioxidant mechanism. *Antioxidants*. 2019. V. 10. P. 1-29. DOI: 10.5772/intechopen.85270.
9. **Koshelev V.N., Primerova O.V., Vorobyev S.V., Ivanova L.V.** Synthesis, redox properties and antibacterial activity of hindered phenols linked to heterocycles. *Molecules*. 2020. V. 25. N 10. P. 2370. DOI: 10.3390/molecules25102370.
10. **Primerova O., Koshelev V., Sabitov E.** Synthesis and antioxidant properties of novel 1, 2, 4-triazoles with 2, 6-di-tert-butylphenol fragments. *IOP Conf. Ser.: Earth Environ. Sci.* 2021. 808. 012030. DOI: 10.1088/1755-1315/808/1/012030.
11. **Yakan H., Omer H.-H., Buruk O., Çakmak Ş., Marah S., Veyisoğlu A., Muğlu H., Ozen T., Kütük H.** Synthesis, structure elucidation, biological activity, enzyme inhibition and molecular docking studies of new Schiff bases based on 5-nitroisatin-thiocarbohydrazone. *J. Mol. Struct.* 2023. V. 1277. P. 134799. DOI: 10.1016/j.molstruc.2022.134799.
12. **Bakır T.K., Lawag J.B.** Preparation, characterization, antioxidant properties of novel Schiff bases including 5-chloroisatin-thiocarbohydrazone. *Res. Chem. Intermed.* 2020. V. 46. P. 2541-2557. DOI: 10.1007/s11164-020-04105-y.
13. **Çavuş M.S., Yakan H., Muğlu H., Bakır T.** Novel carbohydrazones including 5-substituted isatin: Synthesis, characterization, and quantum-chemical studies on the relationship between electronic and antioxidant properties. *J. Phys. Chem. Solids*. 2020. V. 140. P. 109362. DOI: 10.1016/j.jpcs.2020.109362.
14. **El-Mahdy K., El-Kazak A., Abdel-Megid M., Seada M., Farouk O.** Synthesis, Characterization and Antimicrobial Activities of Some New Heterocyclic Schiff Bases Derived from Thiocarbohydrazide. *Acta Chim Slov.* 2016. V. 63. N 1. P. 18-25. DOI: 10.1002/chin.200305140.
15. **Bonaccorso C., Marzo T., La Mendola D.** Biological applications of thiocarbohydrazones and their metal complexes: A perspective review. *Pharmaceuticals*. 2019. V. 13. N 1. P. 4. DOI: 10.3390/ph13010004.
16. **Gudzera O.I., Golub A.G., Bdzhola V.G., Volynets G.P., Lukashov S.S., Kovalenko O.P., Kriklivyi I.A., Yaremchuk A.D., Starosyla S.A., Yarmoluk S.M.** Discovery of potent anti-tuberculosis agents targeting leucyl-tRNA synthetase. *Bioorg. Med. Chem.* 2016. V. 24. N 5. P. 1023-1031. DOI: 10.1016/j.bmc.2016.01.028.

17. **Rudrapal M., Satyanandam R.S., Swaroopini T.S., Lakshmi T.N., Jaha S.K., Zaheera S.** Synthesis and antibacterial activity of some new hydrazones. *Med. Chem. Res.* 2013. V. 22. N 6. P. 2840-2846. DOI: 10.1007/s00044-012-0278-5.
18. **Chimenti F., Bizzarri B., Maccioni E., Secci D., Bolasco A., Fioravanti R., Chimenti P., Granese A., Carradori S., Rivanera D.** Synthesis and in vitro activity of 2-thiazolylhydrazone derivatives compared with the activity of clotrimazole against clinical isolates of Candida spp. *Bioorg. Med. Chem. Lett.* 2007. V. 17. N 16. P. 4635-4640. DOI: 10.1016/j.bmcl.2007.05.078.
19. **Aly A.A., Brown A.B., Abdel-Aziz M., Abuo- Rahma G.E.D.A., Radwan M.F., Ramadan M., Gamal- Eldeen A.M.** Synthesis of new 4-oxo-thiazolidine-5-ylidenes of antitumor and antioxidant activities. *J. Heterocycl. Chem.* 2010. V. 47. N 3. P. 547-554. DOI: 10.1002/chin.201042119.
20. **Hamama W.S., Ibrahim M.E., Ghaith E.A., Zoorob H.H.** Peculiar reaction behavior of 1, 3-oxathiolan-5-one toward various reagents: Molecular modeling studies and in vitro antioxidant and cytotoxicity evaluation. *Synth. Commun.* 2017. V. 47. N 6. P. 566-580. DOI: 10.1080/00397911.2016.1276190.
21. **Muğlu H., Çavuş M.S., Bakır T., Yakan H.** Synthesis, characterization, quantum chemical calculations and antioxidant activity of new bis-isatin carbohydrazone and thiocarbohydrazone derivatives. *J. Mol. Struct.* 2019. V. 1196. P. 819-827. DOI: 10.1016/j.molstruc.2019.07.002.
22. **Komendantova A.S., Scherbakov A.M., Komkov A.V., Chertkova V.V., Gudovanniy A.O., Chernoburova E.I., Sorokin D.V., Dzichenka Y.U., Shirinian V.Z., Volkova Y.A.** Novel steroidal 1, 3, 4-thiadiazines: Synthesis and biological evaluation in androgen receptor-positive prostate cancer 22Rv1 cells. *Bioorg. Chem.* 2019. V. 91. P. 103142. DOI: 10.1016/j.bioorg.2019.103142.
23. **Ragab F.A., Abdel-Aziz S.A., Kamel M., Ouf A.M.A., Allam H.A.** Design, synthesis and biological evaluation of some new 1, 3, 4-thiadiazine-thiourea derivatives as potential antitumor agents against non-small cell lung cancer cells. *Bioorg. Chem.* 2019. V. 93. P. 103323. DOI: 10.1016/j.bioorg.2019.103323.
24. **Gerasimova E., Gazizullina E., Igdisanova D., Sidorova L., Tseitler T., Emelianov V., Chupakhin O., Ivanova A.** Antioxidant properties of 2, 5-substituted 6 H-1, 3, 4-thiadiazines promising for experimental therapy of diabetes mellitus. *Russ. Chem. Bull.* 2022. V. 71. N 12. P. 2730-2739. DOI: 10.1021/acs.chemrestox.8b00175.s001.
25. **Sathyanarayana R., Poojary B., Chandrashekarappa R.B., Kumar H., Merugumolu V.K.** Novel [1, 2, 4] triazolo [3, 4-b][1, 3, 4] thiadiazine derivatives embedded with benzimidazole moiety as potent antioxidants. *J. Chin. Chem. Soc.* 2020. V. 67. N 8. P. 1501-1516. DOI: 10.1002/jccs.201900452.
26. **Li G., Shi Z., Li X., Zhao Z.** Synthesis of new ferrocene bis thiocarbazones under solvent-free conditions using microwave. *J. Chem. Res.* 2011. V. 35. N 5. P. 278-281. DOI: 10.3184/174751911x13043447062703.
27. **Yakan H., Bakır T.K., Çavuş M.S., Muğlu H.** New β-isatin aldehyde-N, N′-thiocarbohydrazones: preparation, spectroscopic studies and DFT approach to antioxidant characteristics. *Res. Chem. Intermed.* 2020. V. 46. P. 5417-5440. DOI: 10.1007/s11164-020-04270-0.
28. **MuĞlu H., Yakan H., Bakir T.K.** Synthesis, spectroscopic studies, and antioxidant activities of novelthio/carbohydrazones and bis-isatin derivatives from terephthalaldehyde. *Turk. J. Chem.* 2020. V. 44. N 1. P. 237-248. DOI: 10.3906/kim-1910-13.
29. **Pretsch E., Bhuhlmann P., Affolter C.** Structure determination of organic compounds. Berlin Heidelberg: Springer-Verlag. 2000. 2000. XV. 421 p. DOI: 10.1007/978-3-662-04201-4.
30. **Tashtoush H.I.T.H.I., Al-Talib M., Shkoor M., Ababneh B., Maslat A.** Cyclization Reactions of Mono-Thiocarbohydrazones with α-Haloketones: Synthesis and Potential Biological Activities of Substituted 1, 3-thiazoles and 1, 3, 4-thiadiazines. *Jordan J.Chem.* 2021. V. 16. N 2. P. 49-57. DOI: 10.47014/16.2.1.
31. **Chunduru V.S.R., Vedula R.R.** One-Pot Synthesis of 1, 3, 4-Thiadiazin-5-yl-chromen-2-one Derivatives via Three-Component Reaction. *Synth. Commun.* 2012. V. 42. N 10. P. 1454-1460. DOI: 10.1080/00397911.2010.540697.
32. **Shkoor M., Al-Abade A., Aleteiwib I., Al-Talib M., Tashtoush H.** Unusual product from the acid-catalyzed one-pot, multicomponent reaction of thiocarbohydrazide, aldehydes, and phenacyl bromides. *Synth. Commun.* 2017. V. 47. N 16. P. 1471-1477. DOI: 10.1080/00397911.2017.1332225.
33. **Abdel- Aziem A.** Synthesis and Antimicrobial Activity of Some Novel Thiazoles, 1, 3, 4-Thiadiazines, 1, 3, 4-Thiadiazoles Incorporating Coumarin Moiety. *J. Heterocycl. Chem.* 2015. V. 52. N 1. P. 251-259. DOI: 10.1002/jhet.2390.
34. **Bondet V., Brand-Williams W., Berset C.** Kinetics and mechanisms of antioxidant activity using the DPPH. free radical method. *LWT - Food Sci. Technol.* 1997. V. 30. N 6. P. 609-615. DOI: 10.1006/fstl.1997.0240.
35. **Jakovljević K., Matić I.Z., Stanojković T., Krivokuća A., Marković V., Joksović M.D., Mihailović N., Nićiforović M., Joksović L.** Synthesis, antioxidant and antiproliferative activities of 1, 3, 4-thiadiazoles derived from phenolic acids. *Bioorg. Med. Chem. Lett.* 2017. V. 27. N 16. P. 3709-3715. DOI: 10.1016/j.bmcl.2017.07.003.

Поступила в редакцию 17.05.2023
Принята к опубликованию 27.09.2023

Received 17.05.2023
Accepted 27.09.2023