ISSN 1070-3632, Russian Journal of General Chemistry, 2022, Vol. 92, No. 2, pp. 166–173. © The Author(s), 2022. This article is an open access publication.
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Synthesis and Antimicrobial Activity of Novel Hydrazone and 1,2,4-Triazole-3-thione Derivatives

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Received November 12, 2021; revised November 12, 2021; accepted November 28, 2021

Abstract—Novel hydrazone and 1,2,4-triazole-3-thione derivatives were obtained via the reaction of N^1,N^3,2-triaryl-6-hydroxy-6-methyl-4-oxocyclohexane-1,3-dicarboxamides with acid hydrazides and thiosemicarbazide, respectively. Structure of the products was proved using IR and ^{1}H NMR spectroscopy methods. Some of the synthesized compounds were tested for antimicrobial activity

Keywords: acid hydrazides, thiosemicarbazide, hydrazones, 1,2,4-triazole-3-thiones, antimicrobial activity

DOI: 10.1134/S1070363222020050

The synthesis and determination of the practical value of hydrazones are relevant, since a hydrazone fragment is present in the molecules of a number of biologically active compounds [1], which have antimicrobial [2–5], anti-inflammatory [6], analgesic [7], antiprotozoal [8], antituberculous [9], anticonvulsant [10], and cardioprotective activity [11]. The combination of different functional groups in hydrazones results in a large number of compounds with unique physical and chemical properties. Some of them can be used in the treatment of diseases of the central nervous system [12], as well as in molecular targeted therapy of drug treatment of cancer [13, 14]. Structural analogs of hydrazones have shown good results in their study as growth promoters in plants of *Nicotiana tabacum L*. and *Arabidopsis thaliana* species [15].

The 1,2,4-triazole-3-thione fragment occurs in the structure of many natural and biologically active compounds [16, 17], for example, in bicyclic anxiolytic drugs, estazolam, alprazolam (Scheme 1), in the triptan 5-HT1 agonist (rizatriptan) and in antimicrobial agents based on spiropiperidinyl-1,2,4-triazolidine-3-thione [18–22]. For the synthesis of heterocyclic compounds with antimicrobial activity with a 1,2,4-triazole-3-thione moiety, the reaction of ketones with thiosemicarbazide is used [23–27].

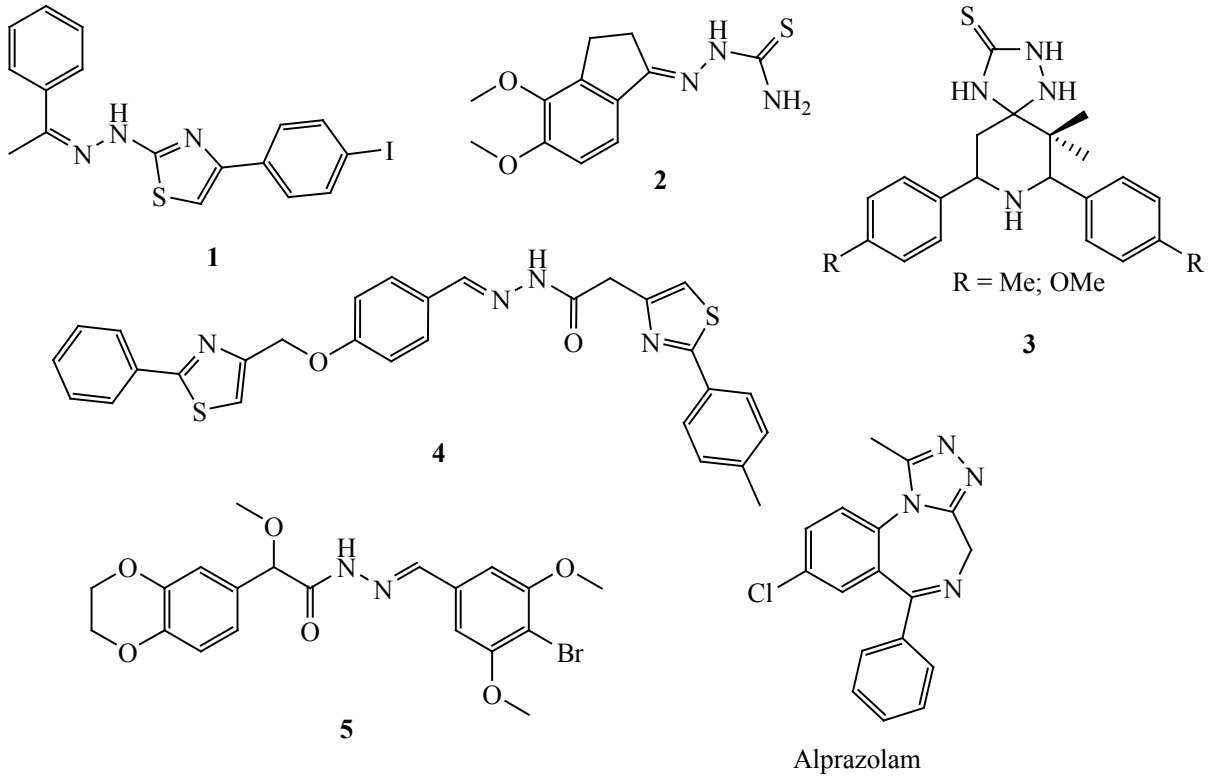
In this regard, the synthesis of compounds with hydrazone and 1,2,4-triazole-3-thione fragments is promising for the preparation of biologically active compounds and for the creation of new drugs based on them.

Scheme 1 shows examples of biologically active hydrazones and 1,2,4-triazole-3-thione derivatives, which have antimicrobial (**1**) [4], antiprotozoal (**2**) [8], antimicrobial (**3**) [21], and anti-inflammatory activity (**4**) [6]. Hydrazone **5** inhibits the phosphodiesterase 10A enzyme responsible for neurological and psychological disorders (schizophrenia) [14].

Previously, we have obtained new oxocyclohexane-1,3-dicarboxamide derivatives by the condensation of acetoacetic acid amides with aromatic aldehydes in the presence of a basic piperidine catalyst in ethanol at room temperature [28–31]. The reactions of the obtained compounds with *N*-nucleophiles [29] and Baeyer–Villiger oxidation [33] have been studied.

In continuation of studying the reactivity of cyclohexanone derivatives [28–34] and in order to obtain new compounds with potential biological activity, herein we reported the reactions of N^1,N^3,2-triaryl-6-hydroxy-6-methyl-4-oxocyclohexane-1,3-dicarboxamides with acid hydrazides and thiosemicarbazide. New derivatives

Scheme 1.



of hydrazones and 1,2,4-triazole-3-thione were obtained, respectively. The reaction of equimolar amounts of 4-oxocyclohexane-1,3-dicarboxamides **6a**–**6l** with hydrazides of salicylic, isonicotinic, and *p*-toluenesulfonic acids upon boiling in ethanol proceeds at the carbonyl group of the alicycle to form the corresponding hydrazones **7a**–**7d**, **8a**, **8b**, **9a**–**9f** (Scheme 2).

The hydrazone form of compounds **7**–**9** is confirmed by the presence in the NMR spectra of the spin-spin coupling between the protons at the C^3 (3.65–4.42 ppm) C^2 atoms of the ring (3.12–4.04 ppm). The proton signal of the NH group not linked to the benzene ring also proves the proposed structure. The chemical shifts of the proton singlets of the two NH groups of the arylamide substituents are shifted to a stronger field compared to the chemical shifts of the starting compounds **6a**–**6o** [28–30].

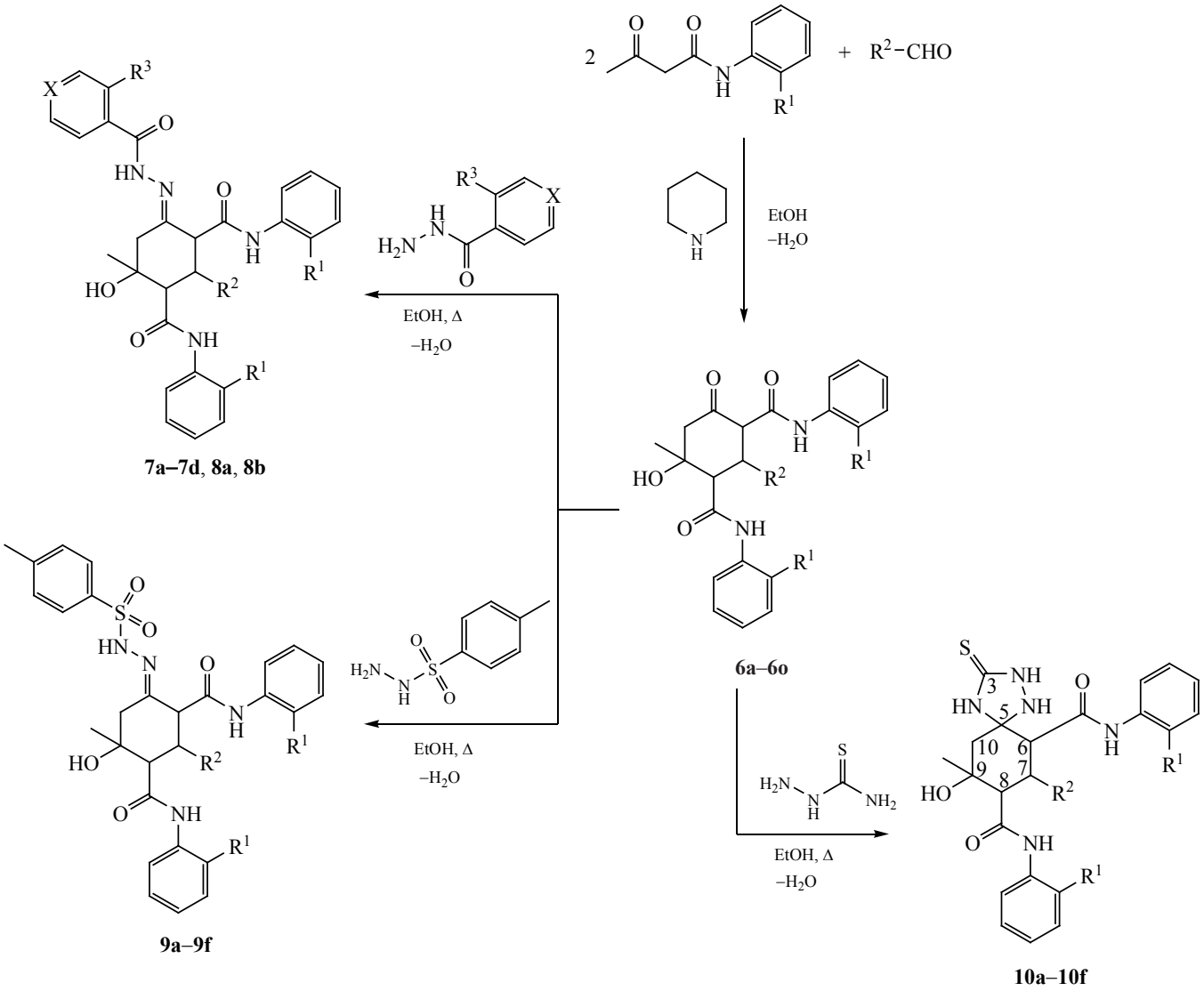
Existence of compounds **7**–**9** in the hydrazone form can be explained by its stabilization due to intermolecular hydrogen bonds. Heterocyclization apparently does not proceed due to the low nucleophilicity of nitrogen atoms in acid hydrazides.

The reaction of cyclohexanone derivatives **6d**, **6m**–**6o** with thiosemicarbazide in an equimolar ratio under similar conditions gave *N*^{6},*N*^{8},7-triaryl-9-hydroxy-9-methyl-3-thioxo-1,2,4-triazaspiro[4.5]decane-6,8-dicarboxamides **10a**–**10d**.

In the IR spectra of compounds **10a**–**10d** there are no stretching vibrations of the conjugated CO group of the alicycle and C=C bonds. The presence of stretching vibration bands of the N(C=S)N fragment at 1336–1360 cm^{-1} and the C=S moiety at 1592–1600 cm^{-1} , as well as the presence of proton signals of the NH groups at the C^1 and C^2 atoms (8.05–8.54 ppm), C^4 (10.33–10.46 ppm) and C^6 atoms (3.54–4.46 ppm) resonating with a proton at the C^7 atom (3.67–4.00 ppm) in the ^{1}H NMR spectra confirm the proposed structure of spiro compounds **10a**–**10d** and excludes possible alternative enamine and imine structures. When comparing the spectral characteristics of spiro compounds **10a**–**10d** with the starting cyclohexanones **6d**, **6m**–**6o**, it was found that the chemical shift of the proton doublet at the C^8 atom of the ring in the spectra of compounds **10a**–**10d** is shifted to upfield region (2.78–3.12 ppm, *J* 11.4–12.0 Hz) [29, 30].

Compounds **7b**, **8b**, **9c**, **10a**, and **10c** were studied for antimicrobial activity against strains of gram-negative (*Escherichia coli* ATCC 25922) and gram-positive

Scheme 2.



R^1 = H, R^2 = 4-BrC_6H_4, R^3 = OH, X = CH (**6a**, **7a**); R^1 = Me, R^2 = 4-ClC_6H_4, R^3 = OH, X = CH (**6b**, **7b**); R^1 = Me, R^2 = pyridin-3-yl, R^3 = OH, X = CH (**6c**, **7c**); R^1 = Cl, R^2 = 4-$Me_2NC_6H_4$, R^3 = OH, X = CH (**6d**, **7d**); R^1 = H, R^2 = 3,4-$(MeO)_2C_6H_3$, R^3 = H, X = N (**6e**, **8a**); R^1 = MeO, R^2 = 4-MeC_6H_3, R^3 = H, X = N (**6f**, **8b**); R^1 = H, R^2 = Ph (**6g**, **9a**), 4-$Me_2NC_6H_4$ (**6h**, **9b**), 4-$Et_2NC_6H_4$ (**6i**, **9c**); R^1 = MeO, R^2 = Ph (**6j**, **9d**), 4-*i*-PrC_6H_4 (**6k**, **9e**), 4-$Me_2NC_6H_4$ (**6l**, **9f**); R^1 = H, R^2 = 4-$EtOC_6H_4$ (**6m**, **10a**); R^1 = MeO, R^2 = thien-2-yl (**6n**, **10b**), pyridin-3-yl (**6o**, **10c**); R^1 = Cl, R^2 = 4-$Me_2NC_6H_4$ (**6d**, **10d**).

bacteria (*Staphylococcus aureus* ATCC 6538-P), as well as *Candida* fungi (*Candida albicans* NCTC 885-653). Minimum inhibitory concentrations (MICs) have been established and range from 500 to 1000 μg/mL (Table 1).

In summary, new derivatives of hydrazones and 1,2,4-triazole-3-thione were obtained by the reaction of substituted 6-hydroxy-6-methyl-4-oxocyclohexane-1,3-dicarboxamides with acid hydrazides and thiosemicarbazide.

EXPERIMENTAL

IR spectra were recorded on a Specord M-80 instrument from KBr pellets. ^{1}H NMR spectra were registered on a Bruker DRX 500 (500 MHz) and Bruker AVANCE III HD 400 (400 MHz) spectrometers in DMSO-d_6,

Table 1. Antimicrobial activity of compounds **7b**, **8b**, **9c**, **10a**, **10c**

Compound	MIC, μg/mL		
	Escherichia coli ATCC 6538-P	*Staphylococcus aureus* ATCC 25922	*Candida albicans* NCTC 885-653
7b	1000	1000	1000
8b	1000	1000	1000
9c	>1000	>1000	1000
10a	500	500	1000
10c	500	500	1000
Furacilin	250	125	–
Dioxidine	62.5–1000	3.9–62.5	–
Fluconazole	–	–	8–31

tetramethylsilane was used as the internal standard. Mass spectra were taken on an ultra-HPLC-MS spectrometer (Waters Acquity UPLC BEH C18 column 1.7 μm, mobile phases were acetonitrile and water, flow rate was 0.6 mL/min, ESI detector MS Xevo TQD). Elemental analysis was performed on an elemental analyzer Euro EA3028-NT for simultaneous determination of C, H, N. Melting points were determined on a Melting Point M-565.

N^1,N^3,2-Triaryl-6-hydroxy-6-methyl-4-oxocyclohexane-1,3-dicarboxamides **6a**–**6o** were prepared according to known methods [28–30].

General procedure for the synthesis of compounds 7–10. A mixture of 0.005 mol of *N*,*N*′,2-triaryl-6-hydroxy-6-methyl-4-oxocyclohexane-1,3-dicarboxamide and 0.005 mol of salicylic (**7a**–**7d**), isonicotinic (**8a**, **8b**), and *p*-toluenesulfonic hydrazides acid (**9a**–**9f**) or thiosemicarbazide (**10a**–**10d**) in 25 mL of ethanol was refluxed for 2.5 h, then cooled. The precipitated crystals were filtered off and crystallized from ethanol.

2-(4-Bromophenyl)-6-hydroxy-4-[2-(2-hydroxyphenyl)hydrazinylidene]-6-methyl-*N*^{1},*N*^{3}-diphenylcyclohexane-1,3-dicarboxamide (7a). Yield 56%, mp 200–201°C. ^{1}H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 1.33 s (3H, Me), 2.35 d (1H, $C^5\underline{H}^AH^B$, *J* 14.0 Hz), 3.01 d (1H, $C^5H^A\underline{H}^B$, *J* 14.0 Hz), 3.31 d (1H, C^1H, *J* 12.0 Hz), 3.75 d (1H, C^3H, *J* 12.0 Hz), 3.88 t (1H, C^2H, *J* 12.0 Hz), 4.94 s (1H, OH), 6.77–7.87 m (18H, $2C_6H_5$, $2C_6H_4$), 9.55 s (1H, C^1CONH), 9.66 s (1H, C^3CONH), 11.21 br. s (2H, 2-$\underline{HO}C_6H_4C\underline{ONH}$). Found, %: C 63.34; H 4.87; N 8.81. $C_{33}H_{31}BrN_4O_4$. Calculated, %: C 63.16; H 4.98; N 8.93.

6-Hydroxy-4-[2-(2-hydroxyphenyl)hydrazinylidene]-6-methyl-*N*^{1},*N*^{3}-di(2-methylphenyl)-2-(4-chlorophenyl)cyclohexane-1,3-dicarboxamide (7b). Yield 32%, mp 245–247°C. ^{1}H NMR spectrum (500 MHz, DMSO-d_6), δ, ppm: 1.40 s (3H, CH_3), 1.80 s (6H, (2-$\underline{Me}C_6H_4)_2$), 2.05 d (1H, $C^5\underline{H}^AH^B$, *J* 14.0 Hz), 2.48 d (1H, $C^5H^A\underline{H}^B$, *J* 14.0 Hz), 3.10 d (1H, C^1H, *J* 12.0 Hz), 3.70 t (1H, C^2H *J* 12.0 Hz), 3.90 d (1H, C^3H, *J* 12.0 Hz), 5.10 s (1H, OH), 6.88–7.30 m (16H, $4C_6H_4$), 7.86 s (1H, C^1CONH), 9.18 s (1H, C^3CONH), 11.15 s (1H, C^4=N$\underline{NH}$CO), 11.70 s (1H, 2-$\underline{HO}C_6H_4$). Found, %: C 67.62; H 5.50; N 8.75. $C_{36}H_{35}ClN_4O_5$. Calculated, %: C 67.65; H 5.52; N 8.77.

6-Hydroxy-4-[2-(2-hydroxyphenyl)hydrazinylidene]-6-methyl-*N*^{1},*N*^{3}-di(2-methylphenyl)-2-(pyridin-3-yl)cyclohexane-1,3-dicarboxamide (7c). Yield 51%, mp 234–235°C. ^{1}H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 1.41 s (3H, Me), 1.81 s (3H, 2-$\underline{Me}C_6H_4$), 1.82 s (3H, 2-$\underline{Me}C_6H_4$), 2.36 d (1H, $C^5\underline{H}^AH^B$, *J* 14.0 Hz), 2.99 d (1H, $C^5H^A\underline{H}^B$, *J* 14.0 Hz), 3.18 d (1H, C^1H, *J* 12.0 Hz), 3.90 d (1H, C^3H, *J* 12.0 Hz), 3.95 t (1H, C^2H, *J* 12.0 Hz), 5.11 s (1H, OH), 6.76–8.43 m (16H, $3C_6H_4$, Py), 9.06 s (1H, C^1CONH), 9.23 s (1H, C^3CONH), 11.12 s (1H, 2-$HOC_6H_4C\underline{ONH}$), 11.60 br. s (1H, 2-$\underline{OH}C_6H_4$CONH). Found, %: C 70.86; H 6.00; N 12.24. $C_{34}H_{35}N_5O_4$. Calculated, %: C 70.69; H 6.11; N 12.12.

6-Hydroxy-4-[2-(2-hydroxyphenyl)hydrazinylidene]-6-methyl-2-(4-dimethylaminophenyl)-*N*^{1},*N*^{3}-di(2-chlorophenyl)cyclohexane-1,3-dicarboxamide (7d). Yield 53%, mp 203–204°C. ^{1}H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 1.37 s (3H, Me), 2.47 d (1H, $C^5\underline{H}^AH^B$, *J* 14.0 Hz), 2.93 s [6H, 4-$\underline{(Me)_2}NC_6H_4$], 3.04 d (1H, $C^5H^A\underline{H}^B$, *J* 14.0 Hz), 3.69 d (1H, C^1H, *J*

12.0 Hz), 3.85 t (1H, C^2H, *J* 12.0 Hz), 4.09 d (1H, C^3H, *J* 12.0 Hz), 5.49 s (1H, OH), 6.46–7.87 m (16H, $4C_6H_4$), 9.39 s (1H, C^1CONH), 9.41 s (1H, C^3CONH), 11.21 br. s (2H, 2-OHC_6H_4CONH). Found, %: C 63.87; H 5.41; N 10.43. $C_{35}H_{35}Cl_2N_5O_4$. Calculated, %: C 63.64; H 5.34; N 10.60.

6-Hydroxy-4-(2-isonicotinoylhydrazinylidene)-6-methyl-2-(3,4-dimethoxyphenyl-*N*^{1},*N*^{3}-diphenylcyclohexane-1,3-dicarboxamide (8a). Yield 30%, mp 231–232°C. ^1H NMR spectrum (500 MHz, DMSO-d_6), δ, ppm: 1.30 s (3H, Me), 2.48 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 2.93 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 3.10 d (1H, C^1H, *J* 12.0 Hz), 3.58 s (3H, 2-$MeOC_6H_3$), 3.60 s (3H, 2-$MeOC_6H_3$), 3.90 t (1H, C^2H, *J* 12.0 Hz), 4.42 d (1H, C^3H, *J* 12.0 Hz), 4.88 br. s (1H, OH), 6.70–7.50 m (17H, $2C_6H_5$, C_6H_3, Py), 9.48 s (1H, C^1CONH), 9.67 s (1H, C^3CONH), 10.80 s (1H, NH). Found, %: C 67.60; H 5.66; N 11.23. $C_{35}H_{35}N_5O_6$. Calculated, %: C 67.62; H 5.67; N 11.27.

6-Hydroxy-4-(2-isonicotinoylhydrazinylidene)-6-methyl-2-(4-methylphenyl)-*N*^{1},*N*^{3}-di(2-methoxyphenyl)cyclohexane-1,3-dicarboxamide (8b). Yield 30%, mp 231–232°C. ^1H NMR spectrum (500 MHz, DMSO-d_6), δ, ppm: 1.31 s (3H, Me), 2.13 s (3H, 4-MeC_6H_4), 2.35 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 2.84 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 3.12 d (1H, C^1H, *J* 12.0 Hz), 3.69 s (3H, 2-$MeOC_6H_4$), 3.75 s (3H, 2-$MeOC_6H_4$), 4.00 t (1H, C^2H, *J* 12.0 Hz), 4.31 d (1H, C^3H, *J* 12.0 Hz), 5.51 br. s (1H, OH), 6.70–7.11 m (16H, $3C_6H_4$, Py), 8.73 s (1H, C^1CONH), 9.12 s (1H, C^3CONH), 10.78 s (1H, NH). Found, %: C 68.00; H 5.85; N 11.00. $C_{36}H_{37}N_5O_6$. Calculated, %: C 68.02; H 5.87; N 11.02.

6-Hydroxy-6-methyl-4-(2-tosylhydrazinylidene)-*N*^{1},*N*^{3},2-triphenylcyclohexane-1,3-dicarboxamide (9a). Yield 60%, mp 235–236°C. IR spectrum (KBr), ν, cm^{-1}: 3400 (OH), 3342 (CONHAr), 3200 (NH), 1668 (CONHAr), 1552 (NH, C=N), 1344, 1168 (SO_2), 904 (S–N). ^1H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 1.30 s (3H, Me), 2.18 s (3H, 4-$MeC_6H_4SO_2$), 2.49 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 2.81 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 3.18 d (1H, C^1H, *J* 12.0 Hz), 3.86 d (1H, C^3H, *J* 12.0 Hz), 3.97 t (1H, C^2H, *J* 12.0 Hz), 5.03 s (1H, OH), 6.90–7.44 m (19H, $3C_6H_5$, C_6H_4), 9.38 s (1H, C^1CONH), 9.56 s (1H, C^3CONH), 10.12 c (1H, C^4NNHSO_2). Found, %: C 66.62; H 5.68; N 9.08. $C_{34}H_{34}N_4O_5S$. Calculated, %: C 66.87; H 5.61; N 9.17.

6-Hydroxy-6-methyl-2-(4-dimethylaminophenyl)-4-(2-tosylhydrazinylidene)-*N*^{1},*N*^{3}-diphenylcyclohexane-1,3-dicarboxamide (9b). Yield 54%, mp 213–214°C. IR spectrum (KBr), ν, cm^{-1}: 3440 (OH), 3344 (CONHAr), 3200 (NH), 1672 (CONHAr), 1552 (NH, C=N), 1344, 1168 (SO_2), 968 (S–N). ^1H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 1.27 s (3H, Me), 2.18 s (3H, 4-$CH_3C_6H_4SO_2$), 2.48 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 2.72 s (6H, 4-$Me_2NC_6H_4$), 2.90 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 3.11 d (1H, C^1H, *J* 12.0 Hz), 3.65 d (1H, C^3H, *J* 12.0 Hz), 3.78 t (1H, C^2H, *J* 12.0 Hz), 4.80 s (1H, OH), 6.37–7.39 m (18H, $2C_6H_5$, $2C_6H_4$), 9.31 s (1H, C^1CONH), 9.42 s (1H, C^3CONH), 9.50 c (1H, C^4NNHSO_2). Found, %: C 66.38; H 5.94; N 10.60. $C_{36}H_{39}N_5O_5S$. Calculated, %: C 66.14; H 6.01; N 10.71.

6-Hydroxy-6-methyl-4-(2-tosylhydrazinylidene)-*N*^{1},*N*^{3}-diphenyl-2-(4-diethylaminophenyl)cyclohexane-1,3-dicarboxamide (9c). Yield 64%, mp 207–208°C. IR spectrum (KBr), ν, cm^{-1}: 3400 (OH), 3344 (CONHAr), 3232 (NH), 1668 (CONHAr), 1552 (NH, C=N), 1380, 1168 (SO_2), 912 (S–N). ^1H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 0.92 t [6H, 4-$(CH_3CH_2)_2NC_6H_4$, *J* 7.0 Hz], 1.16 s (3H, Me), 1.92 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 2.18 s (3H, 4-$MeC_6H_4SO_2$), 2.74 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 3.12 d (1H, C^1H, *J* 12.0 Hz), 3.13 q [4H, 4-$(CH_3CH_2)_2NC_6H_4$, *J* 7.0 Hz], 3.41 d (1H, C^3H, *J* 12.0 Hz), 3.74 t (1H, C^2H, *J* 12.0 Hz), 4.80 s (1H, OH), 6.39–7.44 m (18H, $2C_6H_5$, $2C_6H_4$), 9.25 s (1H, C^1CONH), 9.41 s (1H, C^3CONH), 10.02 c (1H, C^4NNHSO_2). Found, %: C 67.11; H 6.27; N 10.41. $C_{38}H_{43}N_5O_5S$. Calculated, %: C 66.94; H 6.36; N 10.27.

6-Hydroxy-6-methyl-*N*^{1},*N*^{3}-di(2-methoxyphenyl)-4-(2-tosylhydrazinylidene)-2-phenylcyclohexane-1,3-dicarboxamide (9d). Yield 54%, mp 227–228°C. IR spectrum (KBr), ν, cm^{-1}: 3450 (OH), 3352 (CONHAr), 3240 (NH), 1672 (CONHAr), 1540 (NH, C=N), 1336, 1168 (SO_2), 904 (S–N). ^1H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 1.28 s (3H, Me), 2.19 s (3H, 4-$MeC_6H_4SO_2$), 2.47 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 2.85 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 3.10 d (1H, C^1H, *J* 12.0 Hz), 3.67 s (3H, 2-$MeOC_6H_4$), 3.77 s (3H, 2-$MeOC_6H_4$), 3.86 d (1H, C^3H, *J* 12.0 Hz), 4.04 t (1H, C^2H, *J* 12.0 Hz), 5.32 s (1H, OH), 6.59-8.25 m (17H, $3C_6H_4$, C_6H_5), 8.46 s (1H, C^1CONH), 9.12 s (1H, C^3CONH), 9.99 s (1H, C^4NNHSO_2). Found, %: C 64.69; H 5.62; N 8.24. $C_{36}H_{38}N_4O_7S$. Calculated, %: C 64.46; H 5.71; N 8.35.

6-Hydroxy-2-(4-isopropylphenyl)-6-methyl-*N*^{1},*N*^{3}-di(2-methoxyphenyl)-4-(2-tosylhydrazinylidene) cyclohexane-1,3-dicarboxamide (9e). Yield 58%, mp

234–235°C. IR spectrum (KBr), ν, cm^{-1}: 3460 (OH), 3360 (CONHAr), 3254 (NH), 1660 (CONHAr), 1555 (NH, C=N), 1330, 1168 (SO_2), 910 (S–N). ^1H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 1.08 d (6H, 4-$Me_2CHC_6H_4$, *J* 7.0), 1.26 s (3H, Me), 2.18 s (3H, 4-$MeC_6H_4SO_2$), 2.47 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 2.63 m (1H, 4-$Me_2CHC_6H_4$, *J* 7.0 Hz), 2.73 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 3.10 d (1H, C^1H, *J* 12.0 Hz), 3.51 t (1H, C^2H, *J* 12.0 Hz), 3.66 s (3H, 2-$MeOC_6H_4$), 3.75 s (3H, 2-$MeOC_6H_4$), 4.21 d (1H, C^3H, *J* 12.0 Hz), 5.24 s (1H, OH), 6.58–8.16 m (16H, $4C_6H_4$), 8.48 s (1H, C^1CONH), 9.04 s (1H, C^3CONH), 10.01 s (1H, C^4NNHSO_2). Found, %: C 70.49; H 6.58; N 5.26. $C_{32}H_{36}N_2O_6$. Calculated, %: C 70.57; H 6.66; N 5.14.

6-Hydroxy-6-methyl-2-(4-dimethylaminophenyl)-*N*^{1},*N*^{3}-di(2-methoxyphenyl)-4-(2-tosylhydrazinylidene)cyclohexane-1,3-dicarboxamide (9f). Yield 48%, mp 241–242°C. IR spectrum (KBr), ν, cm^{-1}: 3400 (OH), 3380 (CONHAr), 3304 (NH), 1664 (CONHAr), 1552 (NH, C=N), 1312, 1168 (SO_2), 912 (S–N). ^1H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 1.25 s (3H, Me), 2.17 s (3H, 4-$MeC_6H_4SO_2$), 2.47 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 2.73 s (6H, 4-$Me_2NC_6H_4$), 2.84 d (1H, $C^5H^AH^B$, *J* 14.0 Hz), 3.27 d (1H, C^1H, *J* 12.0 Hz), 3.78 s (3H, 2-$MeOC_6H_4$), 3.94 s (3H, 2-$MeOC_6H_4$), 3.95 d (1H, C^3H, *J* 12.0 Hz), 4.00 t (1H, C^2H, *J* 12.0 Hz), 5.32 s (1H, OH), 6.35–8.28 m (16H, $4C_6H_4$), 8.40 s (1H, C^1CONH), 9.07 s (1H, C^3CONH), 9.97 c (1H, C^4NNHSO_2). Found, %: C 63.73; H 6.14; N 9.92. $C_{38}H_{43}N_5O_7S$. Calculated, %: C 63.94; H 6.07; N 9.81.

9-Hydroxy-9-methyl-3-thioxo-*N*^{6},*N*^{8}-diphenyl-7-(4-ethoxyphenyl)-1,2,4-triazaspiro[4.5]decane-6,8-dicarboxamide (10a). Yield 79%, mp 180–181°C. IR spectrum (KBr), ν, cm^{-1}: 3460 (OH), 3360, 3240, 3200, 3080 (NH), 1664 (CONHAr), 1600 (C=S), 1376 (N–CS–N). ^1H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 1.19 t (3H, 4-$MeCH_2OC_6H_4$, *J* 7.0 Hz), 1.28 s (3H, CH_3), 2.14 d (1H, $C^{10}H^AH^B$, *J* 14.6 Hz), 2.89 d (1H, C^8H, *J* 12.0 Hz), 3.28 d (1H, $C^{10}H^AH^B$, *J* 14.6 Hz), 3.55 d (1H, C^6H, *J* 12.0 Hz), 3.89 q (2H, 4-Me$CH_2OC_6H_4$, *J* 7.0 Hz), 3.91 t (1H, C^7H, *J* 12.0 Hz), 4.87 s (1H, OH), 6.59–7.34 m (14H, $2C_6H_5$, C_6H_4), 8.05 c (1H, N^1H), 8.52 s (1H, N^2H), 9.42 s (1H, C^8CONH), 9.62 s (1H, C^6CONH), 10.46 br. s (1H, N^4H). Mass spectrum, *m/z*: 559 $[M + H]^+$. Found, %: C 64.56; H 5.88; N 12.39. $C_{30}H_{33}N_5O_4S$. Calculated, %: C 64.38; H 5.94; N 12.51. *M* 558.

9-Hydroxy-9-methyl-*N*^{6},*N*^{8}-di(2-methoxyphenyl)-7-(thien-2-yl)-3-thioxo-1,2,4-triazaspiro[4.5]decane-6,8-dicarboxamide (10b). Yield 74%, mp 163–164°C. IR spectrum (KBr), ν, cm^{-1}: 3460 (OH), 3390, 3280, 3180, 3010 (NH), 1676 (CONHAr), 1604 (C=S), 1360 (N–CS–N). ^1H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 1.33 s (3H, Me), 2.14 d (1H, $C^{10}H^AH^B$, *J* 14.8 Hz), 2.78 d (1H, C^8H, *J* 11.4 Hz), 3.23 d (1H, $C^{10}H^AH^B$, *J* 14.8 Hz), 3.54 d (1H, C^6H, *J* 11.4 Hz), 3.90 s (3H, 2-$MeOC_6H_4$), 3.93 s (3H, 2-$MeOC_6H_4$), 4.00 t (1H, C^7H, *J* 11.4 Hz), 5.40 s (1H, OH), 6.71–7.76 m (13H, $2C_6H_5$, thienyl), 8.20 s (1H, N^1H), 8.54 s (1H, N^2H), 9.67 s (1H, C^1CONH), 9.71 s (1H, C^3CONH), 10.33 br. s (1H, N^4H). Mass spectrum, *m/z*: 581 $[M + H]^+$. Found, %: C 57.99; H 5.45; N 11.90. $C_{28}H_{31}N_5O_5S_2$. Calculated, %: C 57.81; H 5.37; N 12.04. *M* 580.

9-Hydroxy-9-methyl-*N*^{6},*N*^{8}-di(2-methoxyphenyl)-7-(pyridin-3-yl)-3-thioxo-1,2,4-triazaspiro[4.5]decane-6,8-dicarboxamide (10c). Yield 81%, mp 188–189°C. IR spectrum (KBr), ν, cm^{-1}: 3380 (OH), 3288, 3240, 3120, 3000 (NH), 1648 (CONHAr), 1592 (C=S), 1336 (N–CS–N). ^1H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 1.42 s (3H, Me), 2.19 d (1H, $C^{10}H^AH^B$, *J* 14.8 Hz), 3.12 d (1H, C^8H, *J* 11.6), 3.46 d (1H, $C^{10}H^AH^B$, *J* 14.8 Hz), 3.89 s (3H, 2-$MeOC_6H_4$), 3.91 s (3H, 2-$MeOC_6H_4$), 3.98 t (1H, C^7H, *J* 11.6), 4.46 d (1H, C^6H, *J* 11.6), 5.12 s (1H, OH), 6.87–7.76 m (12H, $2C_6H_4$, Py), 8.41 s (1H, N^1H), 8.52 s (1H, N^2H), 9.23 s (1H, C^8CONH), 9.26 s (1H, C^6CONH), 10.36 br. s (1H, N^4H). Mass spectrum, *m/z*: 576 $[M + H]^+$. Found, %: C 60.21; H 5.66; N 14.72. $C_{29}H_{32}N_6O_5S$. Calculated, %: C 60.40; H 5.59; N 14.57; *M* 575.

9-Hydroxy-7-(4-dimethylaminophenyl)-9-methyl-3-thioxo-*N*^{6},*N*^{8}-di(2-chlorophenyl)-1,2,4-triazaspiro[4.5]-decane-6,8-dicarboxamide (10d). Yield 71%, mp 190–191°C. IR spectrum (KBr), ν, cm^{-1}: 3410 (OH), 3280, 3250, 3131, 3000 (NH), 1668 (CONHAr), 1552 (C=S), 1330 (N–CS–N). ^1H NMR spectrum (400 MHz, DMSO-d_6), δ, ppm: 1.35 s (3H, Me), 2.16 d (1H, $C^{10}H^AH^B$, *J* 14.6 Hz), 2.81 s (6H, 4-$Me_2NC_6H_4$), 2.96 d (1H, C^8H, *J* 11.6), 3.39 d (1H, $C^{10}H^AH^B$, *J* 14.6 Hz), 3.67 t (1H, C^7H, *J* 11.6), 4.26 d (1H, C^6H, *J* 11.6), 5.45 s (1H, OH), 6.46–7.43 m (12H, $3C_6H_4$), 8.13 s (1H, N^1H), 8.15 s (1H, N^2H), 9.28 s (1H, C^8CONH), 9.33 s (1H, C^6CONH), 10.34 br. s (1H, N^4H). Found, %: C 57.62; H 5.07; N 13.27. $C_{30}H_{32}Cl_2N_6O_3S$. Calculated, %: C 57.41; H 5.14; N 13.39.

Antimicrobial activity of compounds **7b**, **8b**, **9c**, **10a**, and **10c** against *Escherichia coli* ATCC 6538-P, *Staphylococcus aureus* ATCC 25922, and *Candida*

albicans NCTC 885-653 strains was determined by successive dilutions of a solution of the test substances in meat-peptone broth at a bacterial load of 250 000 microbial units per 1 mL of the solution. The minimum inhibitory concentration of the compound, i. e., the maximum dilution leading to complete suppression of the test microbes growth, was taken as the effective dose. As reference drugs, furacilin and dioxidine were used for *Escherichia coli* ATCC 25922 and *Staphylococcus aureus* ATCC 6538-P, fluconazole for *Candida albicans* NCTC 885-653.

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CONFLICT OF INTEREST

No conflict of interest was declared by the authors.

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