ISSN 1070-3632, Russian Journal of General Chemistry, 2020, Vol. 90, No. 9, pp. 1611–1619. © Pleiades Publishing, Ltd., 2020.
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Synthesis and Evaluation of Antibacterial Activity of 1,2,4-Oxadiazole-Containing Biphenylcarboxylic Acids

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Received April 21, 2020; revised April 21, 2020; accepted April 30, 2020

Abstract—A one-pot method for the synthesis of biphenylcarboxylic acids containing 1,2,4-oxadiazole ring in the NaOH–DMSO system was developed. The results of *in vitro* experiments showed that the synthesized compounds exhibit antibacterial activity against susceptible strains of *E. coli* and *S. aureus*.

Keywords: heterocycles, dicarboxylic acid anhydrides, basic catalysis, antimicrobial activity, amidoximes

DOI: 10.1134/S1070363220090042

1,2,4-Oxadiazole derivatives have recently attracted much attention of researchers due to the wide use of these heterocycles both in medicinal chemistry and in materials science [1–5]. For example, 1,2,4-oxadizole-containing drugs such as Ataluren (Duchenne's disease) [6], Azilsartan (treatment of hypertension) [7], Opikapon (Parkinson's disease) [8], Oxolamine (a cough suppressant) [9], Amenamivir (HIV therapy) [10], and Naldemidine (pain reliever) [11] are successfully used in medical practice.

One of the priority areas of medical application of 1,2,4-oxadiazoles is the search for new antibacterial agents, primarily against resistant bacterial strains [12–20]. This is due to the fact that the growth of resistance of pathogenic microorganisms to antibacterial drugs is a global threat of the XXI century [21–26].

Earlier, we have developed an efficient method for the synthesis of 1,2,4-oxadiazoles with different functional peripheries based on the condensation of amidoximes with carboxylic acids [27] or their derivatives [28–31]. Subsequently, this approach was successfully applied to obtain biologically active derivatives of 1,2,4-oxadiazole [32–36] (including those with antimicrobial activity [37]) and synthetic building blocks valuable for drug design [38, 39].

Previously, we have reported the reaction of amidoximes with dicarboxylic acid anhydrides, which makes it possible to obtain 1,2,4-oxadiazole systems containing a carboxyl group in a step-economic way under mild conditions [40]. In particular, the 1,2,4-oxadiazole derivative with 2,2′-biphenylcarboxylic acid was obtained for the first time (Scheme 1).

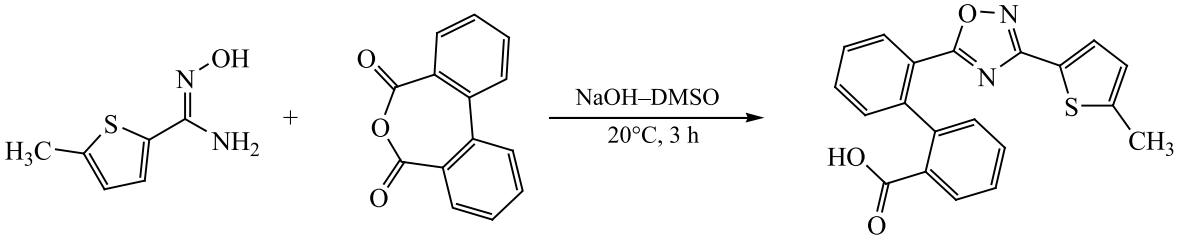
Since 2,2′-biphenyldicarboxylic acids and their derivatives are of interest as potential antimicrobial agents [41–43], we studied the reactions of various aromatic and heterocyclic amidoximes with diphenic anhydrides, and also evaluated antibacterial activity of the resulting hybrid structures against *Staphylococcus aureus* and *Escherichia coli* strains, which are examples of important pathogenic microorganisms.

Amidoximes **2a**–**2h** were obtained by reacting commercially available nitriles **1a**–**1h** with an alcoholic solution of hydroxylamine (Scheme 2) in accordance with the procedure described in [44].

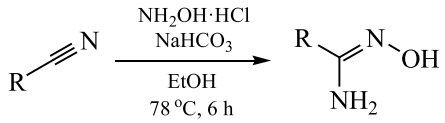
2,2′-Biphenyldicarboxylic acids **4a**–**4c** were synthesized from anthranilic acids **3a**–**3c**, which were first converted to diazonium salts and then subjected to copper-catalyzed coupling (Scheme 3) according to the method described in [45].

Diphenic anhydrides **5a**–**5c** were obtained by dehydration of the corresponding 2,2′-biphenyldicarboxylic acids **4a**–**4c** under the action of trifluoroacetic acid anhydride (TFAA) [46] and reacted *in situ* with amidoximes **2a**–**2h** (Scheme 3). This reaction occurs in two stages: the first is the *O*-acylation of amidoxime **2** with the formation of intermediate **6**, which undergoes

Scheme 1.



Scheme 2.



1a–1h **2a–2h**

R = 4-ClC_6H_4 (**a**, 89%), 4-BrC_6H_4 (**b**, 90%), 4-Py (**c**, 88%), 4-$OCH_3C_6H_4$ (**d**, 78%), 4-$CH_3C_6H_4$ (**e**, 96%), 3-Py (**f**, 86%), 4-$NO_2C_6H_4$ (**g**, 98%), 3-$NO_2C_6H_4$ (**h**, 87%).

then cyclization to 1,2,4-oxadiazole **7** [40]. The second stage of this process is sensitive to the medium pH and can be catalyzed by both acids and bases [47–49]; however, methods using the base catalysis are much more widespread. In our work, for the synthesis of the target 1,2,4-oxadiazoles we used the NaOH–DMSO system, since its effectiveness has been previously proved by the example of reactions of amidoximes with a wide range of carbonyl compounds. Thus, a series of oxadiazole-biphenyl hybrids **7a**–**7j** was obtained in 30–92% yields by the reaction of amidoximes **2a**–**2h** with diphenic anhydrides **5a**–**5c** (Scheme 3).

Antibacterial activity was evaluated by the method of double serial dilutions in accordance with the recommendations [60]. The results are presented in Table 1. A number of compounds **7b**–**7f** did not affect the growth of bacterial cells at concentrations up to 200 μg/mL. Compounds **7g** and **7h** had a bacteriostatic effect at the highest concentration (200 μg/mL). The best results were shown by the acids containing a 4-chlorophenyl substituent at position 3 of the oxadiazole ring. Their bacteriostatic effect was comparable with that of clinically significant antibacterial drugs of the nitrofurans and sulfonamides series (12.5 μg/mL), but significantly lower than the MIC values for the control antibiotic pefloxacin. These compounds (**7a**, **7i**, **7j**) can be recommended as lead compounds for further medicinal chemistry optimization in order to develop a prototype of a new class of antibacterial agents.

Table 1. Antibacterial activity of 1,2,4-oxadiazole derivatives **7a**–**7j**

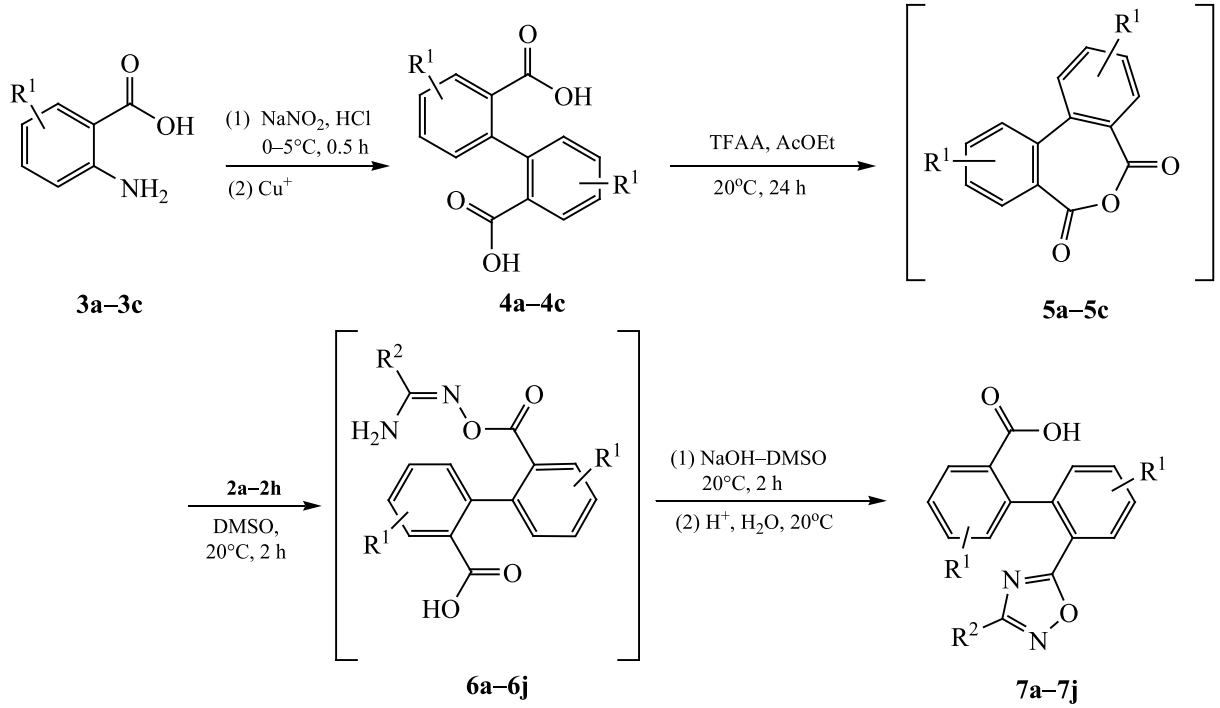
Compound	MIC, μg/mL	
	Staphylococcus aureus (ATCC-25923)	*Escherichia coli* (C600)
7a	50	50
7b	>200	>200
7c	>200	>200
7d	>200	>200
7e	>200	>200
7f	>200	>200
7g	200	200
7h	200	200
7i	>200	12.5
7j	50	12.5
Pefloxacin	<0.5	<0.5

EXPERIMENTAL

Nitriles, substituted anthranilic acids, organic and inorganic reagents and solvents, unless otherwise specified, were obtained from commercial sources (Merck) and were used without further purification. Samples of *S. aureus* reference strains (ATCC-25923) were obtained from the American Type Culture Collection (ATCC); *E. coli* strain (C600) was provided by the Laboratory of Molecular Biology of the G.K. Skryabin Institute of Biochemistry and Physiology of Microorganisms of RAS. For cultivation microorganisms, the nutrient media LB Broth (Lennox) and LB Agar produced by DIA-M (Obninsk) were used.

^1H, ^{13}C, ^{19}F NMR spectra were recorded on a Bruker AVANCE DRX-400 spectrometer with operating

Scheme 3.



R^1 = H, R^2 = 4-ClC_6H_4 (**7a**, 61%), 4-BrC_6H_4 (**7b**, 92%), 4-Py (**7c**, 40%), 4-$CH_3OC_6H_4$ (**7d**, 82%), 4-$CH_3C_6H_4$ (**7e**, 87%), 3-Py (**7f**, 30%), 4-$NO_2C_6H_4$ (**7g**, 35%), 3-$NO_2C_6H_4$ (**7h**, 34%); R^1 = 6,6'-$(CH_3)_2$, R^2 = 4-ClC_6H_4 (**7i**, 81%); R^1 = 5,5'-F_2, R^2 = 4-ClC_6H_4 (**7j**, 72%).

frequencies of 400, 376 and 101 MHz for ^1H, ^{19}F, and ^{13}C, respectively, using DMSO-d_6 or $CDCl_3$ as a solvent. High-resolution mass spectra were recorded on a Bruker Daltonics MicrOTOF-II instrument, ionization method was electrospray (ESI), ionization source temperature was 180°C, eluent was methanol. Melting points were determined on an Electrothermal IA 9300 Series apparatus. The reaction progress was monitored by TLC on Silufol-254 plates (visualization of chromatograms by UV irradiation at 254 nm).

Amidoximes **2a**–**2h** were obtained as described in [44].

***N′*-Hydroxy-4-chlorobenzimidamide (2a)** [44]. Yield 1.51 g (89%), white powder, mp 128–130°C. ^1H NMR spectrum, δ, ppm: 5.85 br. s (2H, NH_2), 7.43 d (2H, Ar, J = 8.6 Hz), 7.66 d (2H, Ar, J = 8.6 Hz), 9.71 s (1H, OH).

***N′*-Hydroxy-4-bromobenzimidamide (2b)** [50]. Yield 1.93 g (90%), beige powder, mp 141–143 °C. ^1H NMR spectrum, δ, ppm: 5.83 br. s (2H, NH_2), 7.55 d (2H, Ar, J = 8.7 Hz), 7.61 d (2H, Ar, J = 8.7 Hz), 9.70 s (1H, OH).

***N′*-Hydroxyisonicotinimidamide (2c)** [51]. Yield 1.13 g (88%), white powder, mp 198–200°C. ^1H NMR spectrum, δ, ppm: 6.00 br. s (2H, NH_2), 7.60–7.69 m (2H, Ar), 8.63–8.52 m (2H, Ar), 10.04 s (1H, OH).

***N′*-Hydroxy-4-methoxybenzimidamide (2d)** [52]. Yield 0.97 g (78%), white powder, mp 107–109°C. ^1H NMR spectrum, δ, ppm: 3.76 s (3H, OCH_3), 5.67 br. s (2H, NH_2), 6.92 d (2H, Ar, J = 7.5 Hz), 7.08 d (2H, Ar, J = 7.5 Hz), 9.43 s (1H, OH).

***N′*-Hydroxy-4-methylbenzimidamide (2e)** [53]. Yield 1.44 g (96%), white powder, mp 141–143°C. ^1H NMR spectrum, δ, ppm: 2.32 s (3H, CH_3), 5.57 br. s (2H, NH_2), 7.15 d (2H, Ar, J = 7.0 Hz), 7.56 d (2H, Ar, J = 7.0 Hz), 9.45 s (1H, OH).

***N′*-Hydroxynicotinimidamide (2f)** [54]. Yield 1.13 g (86%), white powder, mp 107–109°C. ^1H NMR spectrum, δ, ppm: 5.99 br. s (2H, NH_2), 7.33–7.49 m (1H, Ar), 7.81–7.92 m (1H, Ar), 8.40–8.61 m (1H, Ar), 8.85 d (1H, Ar, J = 1.9 Hz), 9.85 s (1H, OH).

***N′*-Hydroxy-4-nitrobenzimidamide (2g)** [55]. Yield 1.20 g (98%), yellow powder, mp 188–190°C. ^1H NMR

spectrum, δ, ppm: 5.94 br. s (2H, NH_2), 7.95 d (2H, Ar, J = 8.5 Hz), 8.2 d (2H, Ar, J = 8.5 Hz), 10.07 s (1H, OH).

***N′*-Hydroxy-3-nitrobenzimidamide (2h)** [56]. Yield 1.57 g (87%), yellow powder, mp 170–172°C. ^{1}H NMR spectrum, δ, ppm: 6.11 br. s (2H, NH_2), 7.69 t (1H, Ar, J = 8.0 Hz), 8.13 d. t (1H, Ar, J = 8.0, 1.0 Hz), 8.23 d. d. d (1H, Ar, J = 8.0, 2.0, 1.0 Hz), 8.52 t (1H, Ar, J = 2.0 Hz), 9.99 s (1H, OH).

[1,1′-Biphenyl]-2,2′-dicarboxylic acids **4a**–**4c** were synthesized according to the procedure [45].

[1,1′-Biphenyl]-2,2′-dicarboxylic acid (4a) [57]. Yield 7.14 g (84%), beige powder, mp 226–228°C. ^{1}H NMR spectrum, δ, ppm (DMSO-d_6): 7.13–7.16 m (2H, Ar), 7.41–7.45 m (2H, Ar), 7.51–7.53 m (2H, Ar), 7.86–7.89 m (2H, Ar), 12.33 br. s (2H, COOH).

6,6′-Dimethyl-[1,1′-biphenyl]-2,2′-dicarboxylic acid (4b) [58]. Yield 1.79 g (97%), beige powder, mp 222–227°C. ^{1}H NMR spectrum, δ, ppm ($CDCl_3$): 2.41 s (6H, CH_3), 7.16–7.21 m (2H, Ar), 7.34–7.38 m (2H, Ar), 7.68–7.71 m (2H, Ar), 11.08 s (2H, COOH).

5,5′-Difluoro-[1,1′-biphenyl]-2,2′-dicarboxylic acid (4c) [59]. Yield 1.50 g (84%), beige powder, mp 245–246°C. ^{1}H NMR spectrum, δ, ppm ($CDCl_3$): 7.14–7.20 m (2H, Ar), 7.39–7.46 m (2H, Ar), 7.64–7.71 m (2H, Ar), 10.56 br. s (2H, COOH).

General procedure for the preparation of (1,2,4-oxadiazol-5-yl)biphenyl-2-carboxylic acids 7a–7j. A solution of trifluoroacetic anhydride (5 mmol) was added with stirring to a suspension of [1,1′-biphenyl]-2,2′-dicarboxylic acid **4** (2.5 mmol) in anhydrous ethyl acetate (20 mL). The reaction mixture was stirred for 24 h, after which the starting material completely dissolved. The resulting solution was concentrated under reduced pressure until a solid residue was formed, which was washed with cold hexane (30 mL) and dried in vacuum. The resulting anhydride was dissolved in DMSO (3 mL) and amidoxime **2** (2.5 mmol) was added. The reaction mixture was stirred at room temperature for 2 h, then powdered sodium hydroxide (5 mmol) was added, and the resulting mixture was stirred for another 1 h. The reaction mixture was diluted with water (30 mL) and acidified with concentrated hydrochloric acid to pH = 1 (for compounds **7a**, **7b**, **7d**, **7e**, **7g**–**7j**), or with acetic acid to pH = 5 (for compounds **7c**, **7f**). The precipitate was filtered off, washed with water (25 mL) and dried in air.

2′-[3-(4-Chlorophenyl)-1,2,4-oxadiazol-5-yl]-biphenyl-2-carboxylic acid (7a). Yield 0.58 g (61%), white powder, mp 160–161°C. ^{1}H NMR spectrum, δ, ppm: 7.29 d (1H, Ar, J = 7.5 Hz), 7.39 d (1H, Ar, J = 7.5 Hz), 7.48–7.67 m (5H, Ar), 7.72 t (1H, Ar, J = 7.4 Hz), 7.87 d (2H, Ar, J = 8.2 Hz), 7.94–8.01 m (1H, Ar), 8.19 d (1H, Ar, J = 7.7 Hz), 12.54 c (1H, COOH). ^{13}C NMR spectrum, δ_C, ppm: 122.8 (Ar), 125.4 (Ar), 128.3 (Ar), 128.4 (Ar), 129.1 (2C, Ar), 129.7 (Ar), 129.8 (2C, Ar), 130.2 (Ar), 131.2 (2C, Ar), 131.2 (Ar), 132.0 (Ar), 132.8 (Ar), 136.8 (Ar), 141.5 (Ar), 143.0 (Ar), 167.2 (oxadiazole), 168.1 (COOH), 176.6 (oxadiazole). Mass spectrum, *m/z*: 399.0519 $[M + Na]^+$ (calcd. $C_{21}H_{13}ClN_2O_3Na$: 399.0507).

2′-[3-(4-Bromophenyl)-1,2,4-oxadiazol-5-yl]biphenyl-2-carboxylic acid (7b). Yield 0.99 g (92%), white powder, mp 150–151°C. ^{1}H NMR spectrum, δ, ppm: 7.29 d. d (1H, Ar, J = 7.6, 1.4 Hz), 7.40 d. d (1H, Ar, J = 7.6, 1.4 Hz), 7.55 t. d (1H, Ar, J = 7.6, 1.4 Hz), 7.63 t. t (2H, Ar, J = 7.6, 1.4 Hz), 7.70–7.82 m (5H, Ar), 7.96 d. d (1H, Ar, J = 7.6, 1.4 Hz), 8.19 d. d (1H, Ar, J = 7.6, 1.4 Hz), 12.56 s (1H, COOH). ^{13}C NMR spectrum, δ_C, ppm: 122.8 (Ar), 125.6 (Ar), 125.8 (Ar), 128.3 (Ar), 128.4 (Ar), 129.3 (2C, Ar), 129.8(Ar), 130.2 (Ar), 131.2 (3C, Ar), 132.0 (Ar), 132.8 (2C, Ar), 132.8 (Ar), 141.5 (Ar), 143.0 (Ar), 167.3 (oxadiazole), 168.1 (COOH), 176.6 (oxadiazole). Mass spectrum, *m/z*: 421.0191 $[M + H]^+$ (calcd. $C_{21}H_{14}BrN_2O_3$: 421.0182).

2′-[3-(Pyridin-4-yl)-1,2,4-oxadiazol-5-yl]biphenyl-2-carboxylic acid (7c). Yield 0.35 g (40%), white powder, mp 206–207°C. ^{1}H NMR spectrum, δ, ppm: 7.30 d. d (1H, Ar, J = 7.6, 1.4 Hz), 7.41 d. d (1H, Ar, J = 7.6, 1.4 Hz), 7.57 t. d (1H, Ar, J = 7.6, 1.4 Hz), 7.65 t. d (2H, Ar, J = 8.1, 1.4 Hz), 7.72–7.76 m (1H, Ar), 7.76–7.83 m (2H, Ar), 7.97 d. d (1H, Ar, J = 7.6, 1.4 Hz), 8.21 d. d (1H, Ar, J = 8.1, 1.4 Hz), 8.81 s (2H, Ar), 12.56 s (1H, COOH). ^{13}C NMR spectrum, δ_C, ppm: 122.6 (Ar), 128.3 (Ar), 128.5 (Ar), 128.6 (Ar), 129.8 (Ar), 130.2 (Ar), 131.2 (Ar), 131.9 (Ar), 132.1 (2C, Ar), 132.1 (Ar) 132.8 (Ar), 133.0 (Ar), 133.8 (Ar), 141.4 (Ar), 143.1 (Ar), 151.4 (2C, Ar), 166.7 (oxadiazole), 168.1 (COOH), 177.1 (oxadiazole). Mass spectrum, *m/z*: 344.1029 $[M + H]^+$ (calcd. $C_{20}H_{14}N_3O_3$: 344.1030).

2′-[3-(4-Methoxyphenyl)-1,2,4-oxadiazol-5-yl]biphenyl-2-carboxylic acid (7d). Yield 0.78 g (82%), white powder, mp 170–171°C. ^{1}H NMR spectrum, δ, ppm: 3.82 s (3H,CH_3), 7.03–7.12 m (2H, Ar), 7.28 d. d (1H, Ar, J = 7.6, 1.4 Hz), 7.38 d. d (1H, Ar, J = 7.6, 1.4 Hz), 7.55 t. d (1H, Ar, J = 7.6, 1.4 Hz), 7.62 t. d (2H, Ar, J = 7.6, 1.4 Hz), 7.71 t. d (1H, Ar, J = 7.6, 1.4 Hz), 7.75–7.83 m (2H, Ar), 7.97 d. d (1H, Ar, J = 7.8, 1.5 Hz),

8.18 d. d (1H, Ar, J = 7.8, 1.5 Hz), 12.49 s (1H, COOH). ^{13}C NMR spectrum, δ_C, ppm: 55.9 (OCH_3), 115.1 (2C, Ar), 118.9 (Ar), 123.0 (Ar), 128.2 (Ar), 128.3 (Ar), 129.0 (2C, Ar), 129.6 (Ar), 130.2 (Ar), 131.1 (Ar), 131.2 (2C, Ar), 132.0 (Ar), 132.6 (Ar), 141.7 (Ar), 143.0 (Ar), 162.2 (Ar), 167.7 (oxadiazole), 168.1 (COOH), 176.0 (oxadiazole). Mass spectrum, *m/z*: 373.1175 $[M + H]^+$ (calcd. $C_{22}H_{17}N_2O_4$: 373.1183).

2′-[3-(4-Methylphenyl)-1,2,4-oxadiazol-5-yl]biphenyl-2-carboxylic acid (7e). Yield 0.79 g (87%), white powder, mp 146–147°C. ^{1}H NMR spectrum, δ, ppm: 2.36 s (3H, CH_3), 7.28 d. d (1H, Ar, J = 7.6, 1.4 Hz), 7.33 d (2H, Ar, J = 7.9 Hz), 7.39 d. d (1H, Ar, J = 7.6, 1.4 Hz), 7.54–7.58 m (1H, Ar), 7.62 t. d (2H, Ar, J = 7.6, 1.4 Hz), 7.71 t. d (1H, Ar, J = 7.6, 1.4 Hz), 7.76 d (2H, Ar, J = 7.8 Hz), 7.97 d. d (1H, Ar, J = 7.6, 1.5 Hz), 8.19 d. d (1H, Ar, J = 7.8, 1.4 Hz), 12.62 s (1H, COOH). ^{13}C NMR spectrum, δ_C, ppm: 21.5 (CH_3), 123.0 (Ar), 123.8 (Ar), 127.3 (2C, Ar), 128.2 (Ar), 128.3 (Ar), 129.7 (Ar), 130.2 (2C, Ar), 130.2 (Ar), 131.1 (Ar), 131.2 (2C, Ar), 132.0 (Ar), 132.6 (Ar), 141.6 (Ar), 141.9 (Ar), 143.0 (Ar), 167.9 (oxadiazole), 168.1 (COOH), 176.2 (oxadiazole). Mass spectrum, *m/z*: 379.1056 $[M + Na]^+$ (calcd. $C_{22}H_{16}N_2O_3Na$: 379.1053).

2′-[3-(Pyridin-3-yl)-1,2,4-oxadiazol-5-yl]biphenyl-2-carboxylic acid (7f). Yield 0.27 g (30%), white powder, mp 205–206°C. ^{1}H NMR spectrum, δ, ppm: 7.30 d. d (1H, Ar, J = 7.6, 1.4 Hz), 7.41 d. d (1H, Ar, J = 7.6, 1.4 Hz), 7.55–7.66 m (4H, Ar), 7.74 t. d (1H, Ar, J = 7.6, 1.4 Hz), 7.98 d. d (1H, Ar, J = 7.7, 1.5 Hz), 8.17–8.27 m (2H, Ar), 8.92 d (2H, Ar, J = 8.6 Hz), 12.57 s (1H, COOH). ^{13}C NMR spectrum, δ_C, ppm: 122.6 (Ar), 127.7 (Ar), 128.3 (Ar), 128.4 (Ar), 129.8 (Ar), 130.2 (Ar), 131.2 (Ar), 131.3 (Ar), 131.3 (Ar), 132.1 (Ar), 132.9 (2C, Ar), 134.8 (Ar), 141.5 (Ar), 143.1 (Ar), 148.2 (Ar), 152.8 (Ar), 166.3 (oxadiazole), 168.1 (COOH), 176.7 (oxadiazole). Mass spectrum, *m/z*: 344.1026 $[M + H]^+$ (calcd. $C_{20}H_{14}N_3O_3$: 344.1030).

2′-[3-(4-Nitrophenyl)-1,2,4-oxadiazol-5-yl]biphenyl-2-carboxylic acid (7g). Yield 0.35 g (35%), beige powder, mp 202–203°C. ^{1}H NMR spectrum, δ, ppm: 7.26–7.34 m (1H, Ar), 7.41 d (1H, Ar, J = 7.6 Hz), 7.57 d (1H, Ar, J = 7.7 Hz), 7.64 t (2H, Ar, J = 7.6 Hz), 7.74 t (1H, Ar, J = 7.6 Hz), 7.97 d (1H, Ar, J = 7.8 Hz), 8.11 d (2H, Ar, J = 8.5 Hz), 8.21 d (1H, Ar, J = 7.8 Hz), 8.35–8.40 m (2H, Ar), 12.62 s (1H, COOH). ^{13}C NMR spectrum, δ_C, ppm: 122.7 (Ar), 124.9 (2C, Ar), 128.3 (Ar), 128.5 (Ar), 128.7 (2C, Ar), 129.9 (Ar), 130.2 (Ar), 131.2 (2C, Ar), 131.3 (Ar), 132.1 (Ar), 132.3 (Ar), 133.0 (Ar), 141.4 (Ar), 143.1 (Ar), 149.7 (Ar), 166.7 (oxadiazole), 168.1 (COOH), 177.1 (oxadiazole). Mass spectrum, *m/z*: 388.0926 $[M + H]^+$ (calcd. $C_{21}H_{14}N_3O_5$: 388.0928).

2′-[3-(3-Nitrophenyl)-1,2,4-oxadiazol-5-yl]biphenyl-2-carboxylic acid (7h). Yield 0.34 g (34%), beige powder, mp 150–151°C. ^{1}H NMR spectrum, δ, ppm: 7.31 d (1H, Ar, J = 7.6 Hz), 7.42 d (1H, Ar, J = 7.6 Hz), 7.57 t (1H, Ar, J = 7.6 Hz), 7.65 t (2H, Ar, J = 7.6 Hz), 7.74 t (1H, Ar, J = 7.6 Hz), 7.84 t (1H, Ar, J = 8.0 Hz), 7.98 d. d (1H, Ar, J = 7.8, 1.4 Hz), 8.22 d. d (1H, Ar, J = 7.8, 1.4 Hz), 8.28 d. t (1H, Ar, J = 7.8, 1.4 Hz), 8.41 d. d (1H, Ar, J = 8.0, 1.0 Hz), 8.53 t (1H, Ar, J = 2.0 Hz), 12.58 s (1H, COOH). ^{13}C NMR spectrum, δ_C, ppm: 122.0 (Ar), 122.6 (Ar), 126.6 (Ar), 128.0 (Ar), 128.3 (Ar), 128.4 (Ar), 129.8 (Ar), 130.2 (Ar), 131.2 (2C, Ar), 131.3 (Ar), 131.6 (Ar), 132.1 (Ar), 133.0 (Ar), 133.3 (Ar), 141.5 (Ar), 143.1 (Ar), 148.7 (Ar), 166.6 (oxadiazole), 168.1 (COOH), 176.9 (oxadiazole). Mass spectrum, *m/z*: 388.0921 $[M + H]^+$ (calcd. $C_{21}H_{14}N_3O_5$: 388.0928).

2′-[3-(4-Chlorophenyl)-1,2,4-oxadiazol-5-yl]-6,6′-dimethyl-[1,1′-biphenyl]-2-carboxylic acid (7i). Yield 0.84 g (81%), white powder, mp 98–100°C. ^{1}H NMR spectrum, δ, ppm: 2.40 s (3H, CH_3), 2.49 s (3H, CH_3), 7.30 t (1H, Ar, J = 7.6 Hz), 7.43–7.59 m (3H, Ar), 7.61–7.65 m (3H, Ar), 7.94 d (1H, Ar, J = 7.7 Hz), 8.11 d (2H, Ar, J = 8.2 Hz), 12.91 s (1H, COOH). ^{13}C NMR spectrum, δ_C, ppm: 20.3 (CH_3), 20.5 (CH_3), 123.9 (Ar), 125.5 (Ar), 127.0 (Ar), 127.8 (Ar), 128.0 (Ar), 129.4 (2C, Ar), 129.9 (2C, Ar), 130.2 (Ar), 131.4 (Ar), 132.9 (Ar), 133.5 (Ar), 135.6 (Ar), 137.0 (Ar), 137.4 (Ar), 137.5 (Ar), 138.6 (Ar), 167.6 (oxadiazole), 167.8 (COOH), 175.2 (oxadiazole). Mass spectrum, *m/z*: 427.0811 $[M + Na]^+$ (calcd. $C_{23}H_{18}ClN_2O_3Na$: 427.0820).

2′-[3-(4-Chlorophenyl)-1,2,4-oxadiazol-5-yl]-5,5′-difluoro-[1,1′-biphenyl]-2-carboxylic acid (7j). Yield 0.75 g (72%), white powder, mp 157–158°C. ^{1}H NMR spectrum, δ, ppm: 7.36–7.39 m (1H, Ar), 7.43–7.56 m (2H, Ar), 7.58–7.63 m (3H, Ar), 7.72 d. d (1H, Ar, J = 9.5, 2.8 Hz), 7.87 d (2H, Ar, J = 8.3 Hz), 7.99 d. d (1H, Ar, J = 9.5, 2.8 Hz), 12.97 s (1H, COOH). ^{13}C NMR spectrum, δ_C, ppm: 116.3 d (J = 24.9 Hz, Ar), 116.8 d (J = 23.2 Hz, Ar), 119.1 d (J = 21.2 Hz, Ar), 119.8 d (J = 21.1 Hz, Ar), 124.5 (Ar), 124.6 (Ar), 125.1 (Ar), 129.1 (2C, Ar), 129.9 (2C, Ar), 133.5 d (J = 7.3 Hz, Ar), 133.7 d (2C, J = 8.0 Hz, Ar), 136.8 d (J = 3.3 Hz, Ar), 137.0 (Ar), 138.3 d (J = 3.4 Hz, Ar), 162.7 d (J = 245.3 Hz, Ar), 163.1 d (J = 245.7 Hz, Ar), 166.8 d (J = 2.0 Hz, oxadiazole), 1.67.3 (COOH), 175.3 d (J = 2.8 Hz, oxadiazole). ^{19}F NMR

spectrum, δ_F, ppm: –114.08, –113.68. Mass spectrum, *m/z*: 413.0498 $[M + H]^+$ (calcd. $C_{21}H_{12}ClF_2N_2O_3$: 413.0499).

Antibacterial activity was evaluated by the double serial dilutions procedure using the turbidimetric method for monitoring the microorganisms growth in accordance with the recommendations [60]. The change in the intensity of light transmission under the influence of solutions of the test substances in the concentration range of 0–200 μg/mL was carried out using a 512 UV/VIS Bibby Scientific Jenway 6715 spectrophotometer. Pefloxacin mesylate (CAS 70458-95-6, Jin Jinle Chemical Co., China) was used as a reference.

Preparation of solutions. The studied drug (5.0 mg) was dissolved in 100 μL of dimethyl sulfoxide, then 10 μL of the resulting solution was taken, and the concentration of the drug was adjusted to 200 μg/mL with sterile LB broth. The formation of a homogeneous solution was observed. A solution of the test compound was introduced into cuvettes ($V = 4$ mL) and a sequential two-fold dilution was made. The resulting dilutions of the drug was as follows: 0.8, 1.6, 3.1, 6.2, 12.5, 25, 50, 100, 200 μg/mL. Sterile cuvettes were prepared with the control of the nutrient medium and placed into a refrigerator, and the cuvettes with the control of the growth of the working suspension (1% control) were placed into an incubatorat 37°C.

Antibacterial screening. The working suspension of the "night" culture of bacteria (0.5 units according to McFarland) in a volume of 100 μL was introduced into the cuvettes, except for the control, which was supplemented with a suspension dilution of 1 : 100 (control 1% of the population). The final volume of the introduced liquid in all cuvettes is 1 mL. After adding all the components, the cuvettes were sealed with sterile tape and the optical density of the resulting suspension was recorded on a spectrophotometer. The cuvettes were incubated for 16–20 h in a thermostat at 35°C. At the end of incubation, the bacterial growth was recorded by the turbidimetric method by changing the optical density of the suspension using a spectrophotometer ($\lambda = 500$ nm). The average values of the optical density of the suspension minus the value of the initial light transmission of the solution (before incubation) in each test cuvette were calculated: (1) negative control of the growth of the working suspension containing the reference antibiotic at a concentration equal to the minimum concentration of the studied substances, (2) 1% control (working suspension, diluted 100 times), (3) each concentration of the test substance. The obtained optical density data was used to plot the dependence of optical density on drug concentration. The minimum inhibitory concentration (MIC) was considered the concentration of the drug at which the average light transmittance of the suspension (based on the results of three experiments) did not significantly exceed 1% of the average value of the growth control and/or the point where the curve reached a plateau.

FUNDING

This work was financially supported by the Russian Foundation for Basic Research (project no. 19-33-60064) using the equipment of the Resource Centers "Chemical Analysis and Materials Research Center" and "Magnetic Resonance Research Center" of the Research Park of St. Petersburg State University.

CONFLICT OF INTEREST

No conflict of interest was declared by the authors.

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