CHEMIJA
2023. Vol. 34. No. 4. P. 171–182

ISSN 0235-7216 eISSN 2424-4538
DOI: https://doi.org/10.6001/chemija.2023.34.4.3

Synthesis and investigation of antibacterial activity of 1-(4-substituted phenyl)-5-oxopyrrolidine-3-carbohydrazide derivatives

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Hydrazides are very important precursors for the synthesis of a wide variety of derivatives, including heterocyclic compounds of biological interest in drug design and discovery. Acid hydrazides with a pyrrole, thiophene, furan, quinoline, isoquinoline, isoxazole and benzimidazole core in the structure were found to be used in medicine, pharmacy, agriculture and many other fields.

In the search for compounds with antibacterial properties, mono- and dihydrazides were used in this study to prepare 1-(4-substituted phenyl)-5-oxopyrrolidine-3-carbohydrazide derivatives bearing the corresponding hydrazone and azole substituents.

The formation of the target compounds was performed via condensation reactions with the chosen aromatic aldehydes and isatin, which led to the construction of the appropriate hydrazone-type structures as well as with diketones which afforded compounds with pyrazole and pyrrole nuclei.

In addition, a preliminary antibacterial evaluation of the synthesised compounds was performed using the gram-positive *Bacillus subtilis* and the gram-negative *Escherichia coli* bacterial strains. As it was expected, the evaluation revealed potential antibacterial candidates that can help address the global challenges of antibiotic resistance and infectious disease outbreaks.

Keywords: 5-oxopyrrolidine, hydrazide, hydrazone, azole, antibacterial activity

INTRODUCTION

Among various synthetic transformations, hydrazides are one of the most attractive precursors for the synthesis of a wide variety of derivatives, including heterocycles [1]. Acid hydrazides include a large group of organic hydrazine derivatives with a functional active group of $C(=O)NHNH_2$. The hydrazinolysis of acyl halides or esters is a common standard pathway for the preparation of hydrazides.

The first representatives – formic and acetic acids hydrazides – were synthesised at the end of the 19th century. The therapeutic importance of acid hydrazides gained momentum with the discovery of isonicotinic acid hydrazide (INH), known as antibiotic isoniazid [2]. It happened in 1952 when its action against *Mycobacterium tuberculosis* was first discovered [3] and has not lost its importance until now [4]. Since then, the modern era of tuberculosis control has begun. Moreover, as revealed later, isonicotinic acid hydrazide inhibits malarial transmission in the mosquito gut [5].

The success of isoniazid led scientists for the design and synthesis of hydrazides with various heterocyclic moieties in their structure, namely furan, pyrrole, thiophene, quinoline, isoquinoline and isoxazole. Their biological assessments showed hydrazides to possess a wide application

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as pharmaceuticals, plant protection agents, substances for manufacturing of polymers, glues, etc., and for many other purposes [6].

In recent years, a number of biologically important hydrazides with various substitutions were synthesised and evaluated for their biological efficacy. They were found to possess antimicrobial and antibacterial [[7–10], antidepressant [11], anti-HCV and anticancer [12, 13], anti-helmintic[14], antioxidant [15], anti-proliferative [16], microbicidal and insecticidal [17] activities. Hydrazides of (acridinyl-9-thio) acetic acid [18] being moderately toxic showed a variety of therapeutic properties, viz. neurotropic, anti-inflammatory, analgesic, anti-microbial and fungistatic effects. 6-Hydroxy-benzofuran-5-carbohydrazide [19] was found to be antifungal. In addition, maleic hydrazide [20] has long been identified as a plant germination inhibitor with growth regulatory and herbicidal effects.

Recently, the World Health Organizations have increasingly announced the increasing resistance to antimicrobial agents, which undoubtedly poses a threat to human health. That is why the discovery of new effective antibacterial drugs has become extremely important. Therefore, our priority area of activity is the synthesis and investigation of hydrazide derivatives with antibacterial properties.

In continuation of our studies to find leading compounds with antibacterial activity [21–23], here we report the synthesis and antibacterial evaluation of some compounds containing mono- and asymmetrical bishydrazone as well as azole cores.

EXPERIMENTAL

Synthesis

Commercially available solvents and reagents were used without further purification unless otherwise mentioned. Reagents and solvents were obtained from Sigma-Aldrich (St. Louis, MO, USA) and used without further purification. The reaction course and purity of the synthesised compounds were monitored by TLC using aluminium plates precoated with Silica gel with F254 nm (Merck KGaA, Darmstadt, Germany). Melting points were determined with a Melt-Temp melting point analyser (Barnstead International, 2555 Kerper Boulevard, Dubuque, Iowa 52001, USA) and were uncorrected. NMR spectra were recorded on a Brucker BioSpin GmbH (400, 101 MHz) spectrometer. Chemical shifts were reported in (δ) ppm relative to tetramethyl silane (TMS) with the residual solvent as internal reference (DMSO-d_6, δ = 2.50 ppm for ^{1}H and δ = 39.5 ppm for ^{13}C). Data were reported as follows: chemical shift, multiplicity, coupling constant (Hz), integration and assignment. IR spectra (ν, cm^{-1}) were recorded on a Bruker TENSOR 27 spectrometer using KBr pellets. Elemental analyses (C, H, N) were conducted using the Elemental Analyzer CE-440; their results were found to be in good agreement (±0.3%) with the calculated values.

GENERAL PROCEDURE OF THE PREPARATION OF HYDRAZONES (2–6)a

A solution of hydrazide **1a** (1 g, 3.6 mmol) and the corresponding aldehyde (3.6 mmol) in a mixture of methanol and 1,4-dioxane (10: 30 mL), in the presence of a catalytic amount of acetic acid, was refluxed for 8–17 h. After completion of the reaction, the mixture was cooled, the formed precipitate was filtered off, washed with diethyl ether and dried.

Methyl 4-(4-(2-(2-methoxybenzylidene) hydrazine-1-carbonyl)-2-oxopyrrolidin-1-yl) benzoate (2a)

White solid, yield 1.23 g (72%), mp 183–185°C (from 1,4-dioxane).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.74–2.93 (m, 2H, $COCH_2$); 3.84; 3.85 (2s, 6H, OCH_3); 3.98–4.23 (m, 3H, NCH_2, CH); 6.94–8.00 (m, 8H, H_{Ar}); 8.38; 8.57 (2s $_{(65:35)}$, 1H, N=CH); 11.55; 11.65 (2s $_{(65:35)}$, 1H, CONH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 32.76 ($CO\underline{CH_2}$); 35.15 (CH); 49.95; (NCH_2); 51.97; 55.68 (OCH_3); 111.79; 111.84; 118.47; 118.52; 120.75; 122.01; 122.12; 124.41; 124.45; 125.44; 125.50; 130.02; 131.40;131.65; 139.43; 142.56; 143.24; 143.31; 157.74 (C_{Ar}); 165.77 (CO); 168.27 (N=C); 172.96; 173.29 (CO) ppm.

IR (KBr): ν, cm^{-1}: 3185 (NH); 1723; 1707; 1664 (3CO); 1283 (OCH_3).

Calcd for $C_{21}H_{21}N_3O_5$, %: C 63.79; H 5.35; N 10.63. Found, %: C 63.82; H 5.51; N 10.52.

Methyl 4-(4-(2-(2,4-dimethoxybenzylidene) hydrazine-1-carbonyl)-2-oxopyrrolidin-1-yl) benzoate (3a)

White solid, yield 0.98 g (69%), mp 202–204°C (from 1,4-dioxane).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.73–2.91 (m, 2H, $COCH_2$); 3.80; 3.83; 3.85 (3s, 9H, OCH_3); 3.97–4.22 (m, 3H, NCH_2, CH); 6.55–8.00 (m, 7H, H_{Ar}); 8.28; 8.47 (2s $_{(65:35)}$, 1H, N=CH); 11.40; 11.50 (2s $_{(65:35)}$, 1H, CONH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 32.76 ($CO\underline{CH_2}$); 35.15 (CH); 49,98 (NCH_2); 51.97; 55.43; 55.76 (OCH_3); 98.13; 98.28; 106.40; 106.48; 114.80; 114.98; 118.46; 118.51; 124.40; 126.66; 130.03; 139.55; 142.66; 143.25; 143.32; 159.02; 159.13; 162.28; 162.50 (C_{Ar}); 165.77 (CO) 167.99 (N=C); 172.78; 172.99 (CO) ppm.

IR (KBr): ν, cm^{-1}: 3 177 (NH); 1707; 1672; 1607 (3CO); 1279 (OCH_3).

Calcd for $C_{22}H_{23}N_3O_6$, %: C 62.11; H 5.45; N 9.88. Found, %: C 62.04; H 5.47; N 9.75.

Methyl 4-(4-(2-(2,5-dimethoxybenzylidene) hydrazine-1-carbonyl)-2-oxopyrrolidin-1-yl) benzoate (4a)

White solid, yield 1.18 g (78%), mp 222–224°C (from 1,4-dioxane).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.75–2.92 (m, 2H, $COCH_2$); 3.72; 3.79; 3.83 (3s, 9H, OCH_3); 3.97–4.24 (m, 3H, NCH_2, CH); 6.95–8.00 (m, 7H, H_{Ar}); 8.34; 8.54 (2s $_{(65:35)}$, 1H, N=CH); 11.56; 11.67 (2s $_{(65:35)}$, 1H, CONH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 32.72 ($CO\underline{CH_2}$); 35.16 (CH); 50.00 (NCH_2); 51.98; 55.41; 56.19 (OCH_3); 109.09; 109.98; 113.22; 113.43; 116.83; 117.71; 118.49; 122.58; 122.77; 124.41; 124.46; 130.03; 139.20; 142.44; 143.24; 143.32; 152.13; 152.27; 153.24 (C_{Ar}); 165.77 (CO); 168.32 (N=C); 172.99; 173.38 (CO) ppm.

IR (KBr): ν, cm^{-1}: 3184 (NH); 1703; 1675; 1606 (3CO); 1291 (OCH_3).

Calcd for $C_{22}H_{23}N_3O_6$, %: C 62.11; H 5.45; N 9.88. Found, %: C 62.05; H 5.48; N 9.76.

Methyl 4-(2-oxo-4-(2-(thiophen-2-ylmethylene) hydrazine-1-carbonyl)pyrrolidin-1-yl)benzoate (5a)

White solid, yield 1.25 g (78%), mp 226–228°C (from 1,4-dioxane).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.72–2.93 (m, 2H, $COCH_2$); 3.83 (s, 3H, OCH_3); 3.96–4.25 (m, 3H, NCH_2, CH); 7.41–8.01 (m, 7H, H_{Ar}); 8.07; 8.26 (2s $_{(65:35)}$, 1H, N=CH); 11.49; 11.53 (2s $_{(65:35)}$, 1H, CONH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 32.69 ($CO\underline{CH_2}$); 35.07 (CH); 49.93 (NCH_2); 51.98 (OCH_3); 118.47; 118.53; 124.42; 124.62; 124.73; 127.56; 127.83; 128.40; 130.03; 137.29; 137.34; 139.44; 142.82; 143.23 143.31 (C_{Ar}); 165.77 (CO); 168.42 (N=C); 172.98; 173.24 (CO).

IR (KBr): ν, cm^{-1}: 3258 (NH); 1716; 1677; 1655 (3CO); 1288 (OCH_3).

Calcd for $C_{18}H_{17}N_3O_4S$, %: C 58.21; H 4.61; N 11.31. Found, %: C 58.12; H 4.68; N 11.25.

Methyl 4-(4-(2-((5-nitrothien-2-yl)methylene) hydrazine-1-carbonyl)-2-oxopyrrolidin-1-yl) benzoate (6a)

Yellow solid, yield 1.12 g (70%), mp 239–241°C (from 1,4-dioxane).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.72–2.94 (m, 2H, $COCH_2$); 3.83 (s, 3H, OCH_3); 3.98–4.23 (m, 3H, NCH_2, CH); 7.51–8.15 (m, 6H, H_{Ar}); 8.20; 8.47 (2s $_{(65:35)}$, 1H, N=CH); 11.99; 12.02 (2s $_{(65:35)}$, 1H, CONH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 32.77 ($CO\underline{CH_2}$); 35.11 (CH); 49.76 (NCH_2); 51.99 (OCH_3); 118.51; 124.46; 129.25; 129.81; 130.04; 130.46; 130.61; 137.00; 140.68; 143.26; 146.45; 146.55; 150.59; 150.90 (C_{Ar}); 165.76 (CO); 169.07 (N=C); 172.75; 173.70 (CO) ppm.

IR (KBr): ν, cm^{-1}: 3222 (NH); 1714; 1692; 1677 (3CO); 1278 (OCH_3).

Calcd for $C_{18}H_{16}N_4O_6S$, %: C 51.92; H 3.87; N 13.45. Found, %: C 51.80; H 3.93; N 13.42.

Methyl 4-(2-oxo-4-(2-(2-oxoindolin-3-ylidene)hydrazine-1-carbonyl)pyrrolidin-1-yl) benzoate (7)

To a solution of hydrazide **1a** (0.5 g, 1.8 mmol) in the mixture of ethanol and 1,4-dioxane (5: 10 mL), in the presence of a catalytic amount of glacial acetic acid, isatin (0.3 g, 2.07 mmol) was added and the mixture was heated at reflux for 13 h. Then the mixture was cooled, and the formed precipitate was filtered off and washed with diethyl ether.

Yellow solid 0.64 g (88%), mp 253–255°C (from methanol).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.76–3.02 (m, 2H, $COCH_2$); 3.83 (s, 3H, OCH_3); 4.01–4.32 (m, 3H, NCH_2, CH); 6.89–8.00 (m, 8H, H_{Ar}); 11.27 (s, 1H, NH); 12.59; 13.08 (2s $_{(65:35)}$, 1H, CONH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 32.24 ($CO\underline{CH_2}$); 34.87 (CH); 49.57 (NCH_2); 51.99 (OCH_3); 111.15; 118.59; 119.75; 120.86; 122.57; 124.53; 130.02; 131,70; 142.52; 143,18 (C_{Ar}); 162.48, 165.75; 172.58; 174.20 (CO) ppm.

IR (KBr): ν, cm^{-1}: 3149 (NH); 1725; 1708; 1688; 1627 (4CO); 1277 (OCH_3).

Calcd for $C_{21}H_{18}N_4O_5$, %: C 62.06; H 4.46; N 13.79. Found, %: C 62.12; H 4.52; N 13.73.

4-(2-Oxo-4-(5-thioxo-4,5-dihydro-1,3,4-oxadiazol-2-yl)pyrrolidin-1-yl)benzoic acid (8)

To a solution of potassium hydroxide (2.02 g, 36 mmol) in methanol (30 mL) carbon disulfide (2.17 mL, 36 mmol) was added dropwise and the mixture was stirred at room temperature for 30 min. Afterwards, hydrazide **1a** (1 g, 3.6 mmol) was added and the mixture was refluxed for 25 h. After completion of the reaction, methanol was evaporated under reduced pressure, and the residue of potassium dithiocarbazate was dissolved in water (20 mL) and refluxed for 4 min with activated carbon. Then it was filtered off and cooled. The filtrate was acetified with hydrochloric acid to pH 1. The formed precipitate was filtered off and washed with water and propan-2-ol.

Light yellow solid, yield 0.759 g (66%), mp 242–243°C (from methanol).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.82–3.09 (m, 2H, $COCH_2$); 3.90–4.32 (m, 3H, NCH_2, CH); 7.79 (d, J = 8.2 Hz, 2H, H_{Ar}); 7.95 (d, J = 8.2 Hz, 2H, H_{Ar}); 12.83 (br. s, 1H, OH); 14.47 (br.s. 1H, NH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 27.85 ($CO\underline{CH_2}$); 35.29 (CH); 49.75 (NCH_2); 118.68; 125.96; 130.16; 142.67 (C_{Ar}); 163.78 (N=C); 166.84; 171.67 (CO); 178.02 (C=S) ppm.

Calcd for $C_{13}H_{11}N_3O_4S$, %: C 51.14; H 3.63; N 13.76. Found, %: C 51.20; H 3.54; N 13.64.

Methyl 4-(4-(2-acetylhydrazine-1-carbonyl)-2-oxopyrrolidin-1-yl)benzoate (9)

To a solution of hydrazide **1a** (1 g, 3.6 mmol) in glacial acetic acid (20 mL) diethylmalonate (0.66 mL, 4.32 mmol) was added dropwise and the mixture was heated at reflux for 24 h. After completion of the reaction (TLC), the mixture was cooled, the obtained precipitate was filtered off and washed with diethyl ether.

White solid, yield 1.02 g (82%), mp 198–199°C (from propan-2-ol).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 1.86 (s, 2H, CH_3); 2.61–2.89 (m, 2H, $COCH_2$); 3.41–3.53 (m, 1H, CH); 3.83 (s, 3H, OCH_3); 3.86–4.15 (m, 2H, NCH_2); 7.82 (d, J = 8.6 Hz, 2H, H_{Ar}); 7.97 (d, J = 8.6 Hz, 2H, H_{Ar}); 9.87 (s, 1H, NH); 10.05 (s, 1H, NH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 20.45 (CH_3); 33.64 ($CO\underline{CH_2}$); 35.79 (CH); 50.43 (NCH_2); 52.00 (OCH_3); 118.47; 124.37; 130.06; 143.20 (C_{Ar}); 165.76; 168.11; 171.30; 172.69 (4CO) ppm.

Calcd for $C_{15}H_{17}N_3O_5$, %: C 56.42; H 5.37; N 13.16. Found, %: C 56.34; H 5.42; N 13.12.

General procedure for the preparation of dihydrazones (10–15)b

A solution of hydrazide **1b** (1.2 g, 4.3 mmol) and the corresponding aldehyde (9.46 mmol) in a mixture of methanol and 1,4-dioxane (10: 30 mL), in the presence of a catalytic amount of acetic acid, was refluxed for 15–21 h. After completion of the reaction, the mixture was cooled, the formed precipitate was filtered off, washed with water and diethyl ether ((**10–12, 14, 15**)**b**) or hexane (**13b**) and dried.

***N*'-(2-methoxybenzylidene)-1-(4-(2-(2-methoxybenzylidene)hydrazine-1-carbonyl) phenyl)-5-oxopyrrolidine-3-carbohydrazide (10b)**

White solid, yield 1.58 g (71%), mp 247–249°C (from 1,4-dioxane).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.75–2.94 (m, 2H, $COCH_2$); 3.85; 3.87 (2s, 6H, OCH_3); 4.00–4.24 (m, 3H, NCH_2, CH); 6.95–8.03 (m, 12H, H_{Ar}); 8.39; 8.58 (2s $_{(65:35)}$, 1H, N=CH); 8.82 (s, 1H, N=CH); 11.56; 11.66; (2s $_{(65:35)}$, 1H, CONH); 11.82 (s, 1H, CONH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 32.81 ($CO\underline{CH_2}$); 35.16 (CH); 50.00; (NCH_2) 55.68 (OCH_3); 111.84; 118.51; 120.76; 122.02; 122.13; 122.40; 125.50; 128.21; 128.39; 131.41; 131.50; 131.66; 139.42; 142.57; 142.94; 157.64; 157.74; 162.20; (C_{Ar}); 168.32 (N=C); 172.52; 172.74; 173.36 (CO) ppm.

IR (KBr): ν, cm^{-1}: 3204 (NH); 1720; 1678; 1639 (3CO); 1251 (OCH_3).

Calcd for $C_{28}H_{27}N_5O_5$, %: C 65.49; H 5.30; N 13.64. Found, %: C 65.42; H 5.25; N 13.71.

_N_'-(2,4-dimethoxybenzylidene)-1-(4-(2-(2,4-dimethoxybenzylidene)hydrazine-1-carbonyl) phenyl)-5-oxopyrrolidine-3-carbohydrazide (11b)

Light yellow solid, yield 1.53 g (74%) mp 184–186°C (from propan-2-ol).

^{1}H BMR (400 MHz) (DMSO–d_6), δ: 2.72–2.92 (m, 2H, $COCH_2$); 3.81; 3.82; 3.84; 3.86 (4s, 12H, OCH_3); 3.97–4.24 (m, 3H, NCH_2, CH); 6.54–8.01 (s, 10H, H_{Ar}); 8.29; 8.48 (2s $_{(65:35)}$, 1H, N=CH); 8.72 (s, 1H, N=CH); 11.41; 11.51 (2s $_{(65:35)}$, 1H, CONH); 11.67 (s, 1H, CONH) ppm.

^{13}C BMR (101 MHz) (DMSO–d_6), δ: 32.80 ($CO\underline{CH_2}$); 35.15 (CH); 50.04 (NCH_2); 55.44; 55.76 (OCH_3); 98.14; 98.29; 106.39; 106.51; 114.81; 114.99; 115.22; 118.42; 118.49; 126.67; 128.30; 139.54; 142.65; 143.05; 159.02; 159.12; 162.00; 162.28; 162.40; 162.51 (C_{Ar}); 168.03 (N=C); 172.53; 172.75; 173.06 (CO).

IR (KBr): ν, cm^{-1}: 3261 (NH); 1678; 1650; 1607 (3CO); 1276 (OCH_3).

Calcd for $C_{30}H_{31}N_5O_7$, %: C 62.82; H 5.45; N 12.21. Found, %: C 62.78; H 5.49; N 12.12.

_N_'-(2,5-dimethoxybenzylidene)-1-(4-(2-(2,5-dimethoxybenzylidene)hydrazine-1-carbonyl) phenyl)-5-oxopyrrolidine-3-carbohydrazide (12b)

White solid, yield 1.79 g (72%), mp 267–269°C (from methanol).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.73–2.94 (m, 2H, $COCH_2$); 3.73; 3.76; 3.80; 3.82 (4s, 12H, OCH_3); 3.99–4.26 (m, 3H, NCH_2, CH); 6.93–8.02 (m, 10H, H_{Ar}); 8.35; 8.55 (2s $_{(65:35)}$, 1H, N=CH); 8.79 (s, 1H, N=CH); 11.57; 11.69 (2s $_{(65:35)}$, 1H, CONH); 11.84 (s, 1H, CONH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 32.76 ($CO\underline{CH_2}$); 35.18 (CH); 50.43 (NCH_2); 55.43; 56.19; 56.23 (OCH_3); 109.17; 109.89; 113.23; 113.41; 116.83; 117.55; 117.71; 118.48; 122.59; 122.79; 123.00; 128.16; 128.40; 139.20; 142.11; 142.45; 142.86; 152.13; 152.28; 152.25 (C_{Ar}); 162.23; 168.38 (N=C); 172.52; 172.77; 173.45 (CO) ppm.

IR (KBr): ν, cm^{-1}: 3210 (NH); 1724; 1678; 1638 (3CO); 1282 (OCH_3).

Calcd for $C_{30}H_{31}N_5O_7$, %: C 62.82; H 5.45; N 12.21. Found, %: C 62.74; H 5.49; N 12.12.

5-Oxo-_N_'-((thien-2-ylmethylene)-1-(4-(2-(thien-2-ylmethylene)hydrazine-1-carbonyl)phenyl) pyrrolidine-3-carbohydrazide (13b)

White solid, yield 1.38 g (69%), mp 268–270°C (from 1,4-dioxane-water mixture).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.73–2.90 (m, 2H, $COCH_2$); 3.94–4.22 (m, 3H, NCH_2, CH); 7.08–8.00 (m, 10H, H_{Ar}); 8.22; 8.44 (2s $_{(65:35:)}$, 1H, N=CH); 8.67 (s, 1H, N=CH); 11.59; 11.62 (2s $_{(65:35:)}$, 1H, CONH); 11.78 (s, 1H, CONH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 32.94 ($CO\underline{CH_2}$); 35.08 (CH); 49.91 (NCH_2); 118.48; 118.52; 127.87; 127.98; 128.22; 128.36; 128.53; 128.89; 129.04; 130.46; 130.85; 131.12; 138.73; 139.21; 142.28; 142.70 (C_{Ar}); 162.26; 168.43 (N=C); 172.50; 172.63; 173.50 (CO) ppm.

IR (KBr): ν, cm^{-1}: 3208 (NH); 1711; 1675; 1638 (3CO).

Calcd for $C_{22}H_{19}N_5O_3S_2$, %: C 56.76; H 4.11; N 15.04. Found, %: C 56.84; H 4.01; N 15.10.

_N_'-((5-nitrothien-2-yl)methylene)-1-(4-(2-((5-nitrothien-2-yl)methylene)hydrazine-1-carbonyl)phenyl)-5-oxopyrrolidine-3-carbohydrazide (14b)

Yellow solid, yield 1.52 g (76%), mp 270–272°C (from 1,4-dioxane).

^{1}H NBMR (400 MHz) (DMSO–d_6), δ: 2.75–2.96 (m, 2H, $COCH_2$); 3.99–4.26 (m, 3H, NCH_2, CH); 7.49–8.17 (m, 8H, H_{Ar}); 8.22; 8.48 (2s $_{(65:35)}$, 1H, N=CH); 8.69 (s, 1H, N=CH); 12.01; 12.04 (2s $_{(65:35:)}$, 1H, CONH); 12.20 (s, 1H, CONH) ppm.

IR (KBr): ν, cm^{-1}: 3278; 3178 (2NH); 1677; 1607 (3CO).

Calcd for $C_{22}H_{17}N_7O_7S_2$, %: C 46.56; H 3.08; N 17.65. Found, %: C 46.41; H 3.15; N 17.56.

_N_'-(4-(dimethylamino)benzylidene)-1-(4-(2-(4-(dimethylamino)benzylidene)hydrazine-1-carbonyl)phenyl)-5-oxopyrrolidine-3-carbohydrazide (15b)

Light yellow solid 1.59 g (69%), mp 273–275°C (from 1,4-dioxane).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.72–2.89 (m, 2H, $COCH_2$); 2.96; 2.97; 298 (3s, 12H, $4CH_3$); 3.96–4.24 (m, 3H, NCH_2, CH); 6.66–7.91 (m, 12H,

H_{Ar}); 7.94; 8.07; (2s $_{(65:35)}$, 1H, N=CH); 8.31 (s, 1H, N=CH); 11.31; 11.36 (2s $_{(65:35)}$, 1H, CONH); 11.51 (s, 1H, CONH) ppm.

IR (KBr): ν, cm^{-1}: 3151 (NH); 1620; 1605; 1572 (CO).

Calcd for $C_{30}H_{33}N_7O_3$, %: C 66.77; H 6.16; N 18.17. Found, %: C 66.65; H 6.24; N 18.25.

5-Oxo-*N*'-(2-oxoindolin-3-ylidene)-1-(4-(2-(2-oxoindolin-3-ylidene)hydrazine-1-carbonyl) phenyl)pyrrolidine-3-carbohydrazide (16). Obtained according to the synthesis method of 7.
Yellow solid, yield 0.675 g (70%), mp 321°C (decomp.) (from propan-2-ol).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.79–3.04 (m, 2H, $COCH_2$); 4.03–4.35 (m, 3H, NCH_2, CH); 6.86–8.10 (m, 12H, H_{Ar}); 11.21; 11.43 (2s, 2H; NH); 12.60; 13.09 (2s $_{(65:35)}$, 1H, CONH); 13.92 (s, 1H, CONH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 32.29 ($CO\underline{CH_2}$); 34.92 (CH); 49.60 (NCH_2); 110.67; 111.18; 111.25; 115.22; 119.03; 119.76; 119.87; 120.95; 121.74; 122.61; 122.85; 128.35; 131.73; 134.86; 142.41; 142.54; 142.87; 142.91 (C_{Ar}); 162.50; 163.11; 172.68; 174.22 (CO) ppm.

IR (KBr): ν, cm^{-1}: 3168 (NH); 1746, 1721, 1701 (3CO).

Calcd for $C_{28}H_{21}N_7O_5$, %: C 62.80; H 3.95; N 18.31. Found, %: C 62.77; H 3.99; N 18.30.

4-(3,5-Dimethyl-1*H*-pyrazole-1-carbonyl)-1-(4-(3,5-dimethyl-1*H*-pyrazole-1-carbonyl)phenyl) pyrrolidin-2-one (17)
A mixture of hydrazide **1b** (3.48 g, 12.55 mmol), pentane-2,4-dione (4.13 g, 41.29 mmol) and ethanol (60 mL) was refluxed for 4 h, then cooled, and the formed solid was filtered off, washed with water.

White solid, yield 3.97 g (78%), mp 238–239°C (from propan-2-ol).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.17, 2.22 (s, 6H, $2CH_3$); 2.49, 2.55 (s, 6H, $2CH_3$); 2.88–3.01 (m, 2H, $COCH_2$); 4.10–4.31 (m, 2H, NCH_2); 4.49–4.56 (m, 1H, CH); 6.25 (s, 1H, CH); 6.28 (s, 1H, CH); 7.83 (d, J = 8.8 Hz, 2H, H_{Ar}); 7.94 (d, J = 8.8 Hz, 2H, H_{Ar}) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 13.86; 13.97, 14.26; 14.43; 35.66; 35.71; 50.37; 111.49; 111.99; 118.26; 128.31; 132.40; 142.83; 144.28; 144.76; 151.81; 152.57 (C_{Ar}); 167.32; 172.76; 172.84 (3CO) ppm.

Calcd for $C_{22}H_{23}N_5O_3$, %: C 65.17; H 5.72; N 17.27. Found, %: C 65.27; H 5.64; N 17.20.

***N*-(2,5-dimethyl-1*H*-pyrrol-1-yl)-1-(4-((2,5-dimethyl-1*H*-pyrrol-1-yl)carbamoyl)phenyl)-5-oxopyrrolidine-3-carboxamide (18).**
A mixture of hydrazide **1b** (3.48 g, 12.55 mmol), hexane-2,5-dione (1.8 g, 15.77 mmol), ethanol (15 mL) and acetic acid (2 mL) was refluxed for 40 h, then the volatile fraction was evaporated under reduced pressure, and the residue was recrystallised from the mixture of water and methanol.

White solid, yield 4.62 g (85%), mp 148–149°C (from water/methanol, 1: 1).

^{1}H NMR (400 MHz) (DMSO–d_6), δ: 2.01 (s, 6H, $2CH_3$); 2.04 (s, 6H, $2CH_3$); 2.77–2.98 (m, 2H, $COCH_2$); 3.47–3.54 (m, 1H, CH); 4.04–4.24 (m, 2H, NCH_2); 5.66 (s, 2H, 2CH); 5.71 (s, 2H, 2CH); 7.88 (d, J = 8.8 Hz, 2H, H_{Ar}); 8.01 (d, J = 8.8 Hz, 2H, H_{Ar}); 10.94 (s, 1H, NH); 11.22 (s, 1H, NH) ppm.

^{13}C NMR (101 MHz) (DMSO–d_6), δ: 10.93, 10.96 ($4CH_3$); 34.00 ($CO\underline{CH_2}$); 35.81 (CH); 50.26 (NCH_2); 103.11; 118.73; 126.73; 126.86; 127.05; 128.31; 142.37 (C_{Ar}); 165.11; 171.74; 172.26 (4CO) ppm.

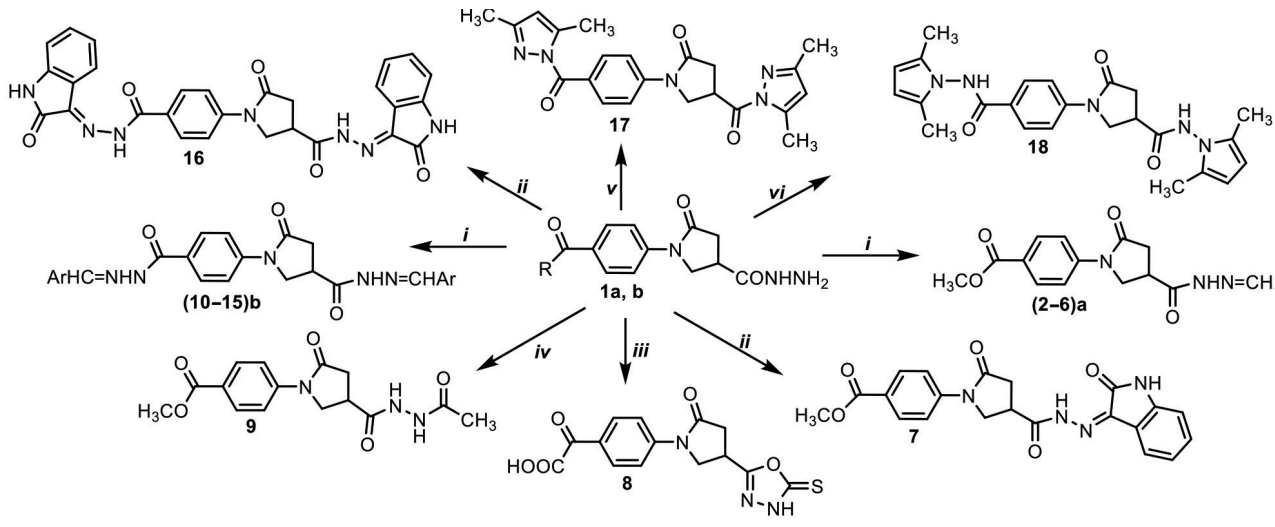
Calcd for $C_{24}H_{27}N_5O_3$, %: C 66.50; H 6.28; N 16.16. Found, %: C 66.42; H 6.36; N 16.05.

RESULTS AND DISCUSSION

Synthesis

The initial compounds **1a, b** having hydrazide functionality were obtained as it is described in Ref. [21]. The synthesised hydrazides **1a, b** with one (**a**) or two (**b**) hydrazinocarbonyl groups were further subjected to various chemical transformations to obtain the target compounds **2–18** (Scheme 1) with hydrazone and azole nuclei in the molecules.

Hydrazones represent a class of organic compounds with a wide range of biological and pharmacological properties. A large number of hydrazones are found to possess a diverse pharmacological activity such as antimicrobial and antifungal, antitumour, central nervous system stabilising, cardio protective, antiviral, antimalarial, analgesic, anti-inflammatory, antiplatelet, anticonvulsant, anthelmintic, antiprotozoal, etc. [24, 25]. That fact led us to the synthesis of a series of hydrazone and dihydrazone derivatives for their antibacterial evaluation.



1a R = OCH_3, 1**b** R = $NHNH_2$;
2, 10 Ar = 2-$H_3COC_6H_4$, **3, 11** Ar = 2,4-di(H_3CO)C_6H_3, **4, 12** Ar = 2,5-di(H_3CO)C_6H_3,
5, 13 Ar = 2-thienyl, **6, 14** Ar = 5-nitro-2-thienyl, **15** Ar = 4-$(H_3C)_2NC_6H_4$

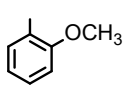
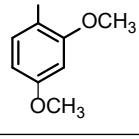
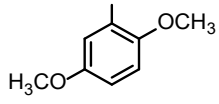
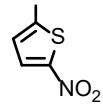
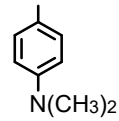
Reagents and conditions: (***i***) the corresponding aromatic aldehyde, CH3OH: 1,4-dioxane, 1:3, glacial CH3COOH, Δ, 8–17 h; (***ii***) isatin, C2H5OH: 1,4-dioxane, 1:2, glacial AcO CH3COOH, Δ, 20 (7) or 32 (16) h; (***iii***) KOH+CH3OH, CS2, stirring 30 min, hydrazide, Δ, 25 h, water, conc. HCl to pH 1; (***iv***) glacial CH3COOH, diethyl malonate, Δ, 24 h; (***v***) pentane-2,4-dione, C2H5OH, Δ, 4 h; (***vi***) hexane-2,5-dione, C2H5OH, glacial CH3COOH, Δ, 40 h.

Scheme 1. Synthesis of compounds **2–18**

Hydrazones are usually obtained through the straightly condensation of carbohydrazides with aldehydes. In this study, to afford monohydrazones **(2–6)a** (Scheme 1), carbohydrazide **1a** was condensed with the appropriate benzaldehydes, namely 2-methoxy-, 2,4-dimethoxy- and 2,5-dimethoxybenzaldehydes, as well as 2-thiophene- and 5-nitro-2-thiophenecarboxaldehydes in a solvent mixture of methanol and 1,4-dioxane (1:3) using a catalytic amount of glacial acetic acid. Depending on the aldehyde used, the reaction time varied between 8 and 17 h. To increase the structural diversity and compound library, synthesis of dihydrazones **(10–15)b** (Scheme 1) was performed. The reactions were carried out analogously to the synthesis of monohydrazones. Dihydrazide **1b** and an excess aromatic aldehyde (1:2.2) were used in the reactions. Depending on the aromatic aldehyde used, the reaction continued for a longer time, i. e. between 15 and 21 h. Aromatic aldehydes used in hydrazone condensation reactions can be seen in Table 1.

The prepared hydrazones were characterised with ^{1}H, ^{13}C NMR and IR spectra, as well as elemental analysis. The NMR spectra of the synthesised hydrazones **(2–6)a** and **(10–15)b** fully proved the formed structures. The azomethine protons of the compounds appeared as two separate singlets in a range of 8.07–8.57 ppm for monohydrazones and in an interval of 7.94–8.58 ppm for di analogues. Furthermore, the proton of the CH=N group of the CONHN=CH fragments which are directly attached to the phenyl ring resonates as a singlet between 8.31 and 8.82 ppm for compounds **(10–15)b**. The amide (NHCO) protons were also observed as two singlets in a range

Table 1. **The yields of the synthesised hydrazones (2–6)a and (10–15)b**

Hydrazones	2a, 10b	3a, 11b	4a, 12b	5a, 13b	6a, 14b	15b
Ar						
Yields, %	72/71	69/74	78/72	78/69	70/76	69

of 11.40–12.02 ppm for monohydrazones **(2–6)a**. The shifts of the analogues signals of the di derivatives **(10–15)b** were almost the same as for mono counterparts and varied between 11.41 and 12.04 ppm. However, for compounds **(10–15)b** the protons of the NHCO group directly attached to the phenyl ring were observed as one singlet in the interval between 11.51 and 12.20 ppm.

As expected, the synthesised hydrazones in DMSO-d_6 solutions exist in the form of two rotamers which is shown by the presence of the splitted signals of amide and azomethine protons.

This splitting is caused by the restricted rotation around the amide bond and shows the presence of a mixture of the *Z/E* rotamers, where the *Z*-form dominates [26, 27]. The *Z* rotamers always resonated up-field of the ^{1}H NMR spectra if compared with the *E*-type. The ^{13}C NMR spectra of the above-mentioned hydrazones also confirm the formation of the desired structures. The carbons of the NHCO gave a shielded rise in the ^{13}C spectra of hydrazones in comparison with the corresponding hidrazides **1a** or **1b**.

The nucleophilic hydrazide group can participate in the reaction with the carbonyl group in the 3rd position of the isatin molecule, and this nucleophilic addition reaction results in hydrazones containing 2-oxoindoline moieties. Isatin hydrazone derivatives exhibit diverse biological activities, including anticancer, antitubercular, antimicrobial and antiviral properties [28, 29].

In the extension of the study, the reaction of monocarbohydrazide **1a** with an equivalent amount of isatin in a mixture of methanol and 1,4-dioxane in the presence of a catalytic amount of glacial acetic acid at reflux for 20 h afforded compound **7**. Meanwhile, compound **16** containing two 2-oxoindoline moieties was obtained by reacting dicarbohydrazide **1b** with excess isatin in a mixture of methanol and 1,4-dioxane for 32 h in the presence of an acidic catalyst. This reaction resulted in 5-oxo-*N*'-(2-oxoindolin-3-ylidene)-1-(4-(2-(2-oxoindolin-3-ylidene)hydrazine-1-carbonyl)phenyl) pyrrolidine-3-carbohydrazide (**16**).

Among the new signals in compound **7**, an NH singlet of the 2-oxoindoline fragment is visible at 11.27 ppm, while the signal of the proton of CONH was seen at 12.59 and 13.08 ppm with the intensity ratio of 65:35 indicating that compound **7** in the DMSO-d_6 solution exists as a mixture of *E/Z* isomers, where the *Z*-form predominates. In the ^{13}C NMR spectrum, due to the isomerisation in DMSO-d_6, the splitting of the resonances of aromatic carbons is observed. The signals of C=O groups were found at the characteristic area of the spectrum, namely at 162.48, 165.75, 172.58 and 174.20 ppm.

Compound **16** containing two 2-oxoindoline moieties shows an additional NH proton signal at 11.43 ppm which belongs to the NH of the newly attached oxoindoline fragment. Like for **7a**, the ^{1}H NMR spectrum of **16** showed an analogous *E/Z* isomerisation due to the presence of an amide bond attached to a pyrrolidinone ring. The proton singlets of this fragment were found to resonate at 12.60 and 13.09 ppm and correspond to signals found in compound **7**. The singlet of the additional CONH which was incorporated at the phenyl ring was found to emerge at 13.92 m. d. and showed no signs of isomerisation. The ^{13}C NMR showed four resonance lines in the lower fields of the spectrum which belong to two oxoindoline, amide and one pyrrolidinone C=O carbons, namely at 162.50, 163.11, 172.68 and 174.22 ppm.

In the course of our study, hydrazides **1a** and **1b** were applied for the formation of azole structural units. This formation is of interest because azoles are known as a class of compounds with excellent therapeutic properties. Azole-based compounds have proved to be a very good source of medicinal agents. Various properties associated with these fragments include antimicrobial, anticancer, antihypertensive, anthelmintic, anti-HIV, anti-inflammatory, analgesic, anticonvulsant, sedative and other pharmacological activities [30]. Therefore, the synthesis of the mentioned compounds is attractive for researches interested in drug discovery and design.

The synthesis of 1,3,4-oxadiazole thione **8** was achieved by the reaction of hydrazide **1a** with carbon disulfide in an alkaline methanol solution. The resulting potassium dithiocarbazate was dissolved in water and heterocyclised with the concentrated hydrochloric acid. However, the presence of the strong acid affected the ester group by hydrolysing it to the acid. Thus, the acidic hydrolysis yielded 4-(2-oxo-4-(5-thioxo-4,5-dihydro-1,3,4-oxadiazol-2-yl)pyrrolidin-1-yl)benzoic acid **(8)** rather than the expected methyl benzoate.

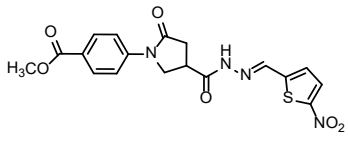
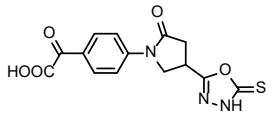
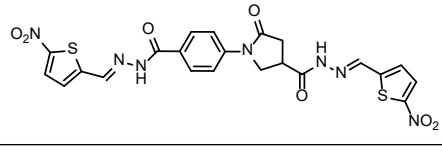
The ^{13}C NMR spectra of 1,3,4-oxadiazole derivative **8** showed the characteristic resonances of the oxadiazole thione ring as follows: 163.78 (N=C), 166.84, 171.67 (2C=O) and 178.02 (C=S) ppm. The presence of a characteristic singlet at 14.47 ppm in the ^{1}H NMR spectrum approved the existing NH in the oxadiazole ring. The broad singlet at 12.83 ppm was assigned to the OH of the COOH fragment resulting due to the acidic hydrolysis.

When reacted with the compounds containing active methylene groups, namely diethyl malonate, hydrazides gave pyrazole dione derivatives. Thus, with hydrazide **1a** in the hand, we targeted to synthesise a compound with a 3,5-dioxopyrazolidin-1-yl fragment in its molecule; however, attempts were unsuccessful. Instead of methyl 4-(4-(3,5-dioxopyrazolidin-1-yl)-2-oxopyrrolidin-1-yl)benzoate, the acetyl product methyl 4-(4-(2-acetylhydrazinyl)-2-oxopyrrolidin-1-yl) benzoate **(9)** was obtained, the structure of which was approved by the data of ^{1}H and ^{13}C NMR spectra. The singlet at 1.86 ppm (^{1}H) and the resonance at 20.45 ppm (^{13}C) were assigned to the protons and the carbon atom of the methyl group, respectively. Two singlets at 9.87 and 10.05 ppm (^{1}H) were ascribed to the NHNH protons, and the spectral lines at 165.76, 168.11, 171.30 and 172.69 ppm (^{13}C) fully correspond to the four carbonyls present in the structure.

And finally, asymmetric bis(3,5-dimethylpyrazole)s and bis (2,5-dimethylpyrrole)s were designed. 4-(3,5-dimethyl-1*H*-pyrazole-1-carbonyl)-1-(4-(3,5-dimethyl-1*H*-pyrazole-1-carbonyl) phenyl)pyrrolidin-2-one **(17)** was obtained from hydrazide **1b** and 3-fold excess pentane-2,4-dione in ethanol at reflux for 4 h. In the ^{1}H NMR spectrum, singlets at 2.17, 2.22, 2.49 and 2.55 ppm correspond to 12 protons of four methyl groups attached to the pyrazole rings. The protons of two CH of the pyrazole cycles were resonated at the characteristic 6.25 and 6.28 ppm. In the ^{13}C NMR, four resonances at 13.86, 13.97, 14.26 and 14.43 ppm, spectral lines at 111.49 and 111.99 ppm, and three resonances at 167.32, 172.76 and 172.84 ppm were assigned to four carbons of the methyl groups, two carbon atoms of the CHs of pyrazoles and three carbons of the C=O groups, respectively, which fully approve the formation of two 3,5-dimethyl-1*H*-pyrazole-1-carbonyl moieties.

The reaction of hydrazide **1b** with an excess hexane-2,5-dione in ethanol at reflux and in the presence of an acidic catalyst resulted in the condensation reaction leading to the formation/introduction of two 2,5-dimethyl pyrrole rings in the scaffold of the desired product **18**. Its ^{1}H NMR spectrum showed the appearance of singlets at 2.01 and 2.04 ppm, which were assigned to the protons of methyl groups, the singlets at 5.66 and 5.71 ppm were attributed to two CH–CH groups of the pyrrole rings, and four aromatic protons were visible as two doublets at 7.88 (J = 8.8 Hz) and 8.01 (J = 8.8 Hz), in

Table 2. **The significant inhibition zones (mm) showed by the synthesised compounds**

Compound	Structural formula	Inhibition zone, mm	
		Gram-negative bacteria	Gram-positive bacteria
		E. coli	*B. subtilis*
6a		18	17
8		–	10
14b		19	18
Ciprofloxacin		30	30

addition signals at 10.94 and 11.22 ppm were attributed to the NH groups of 2,5-dimethyl-1*H*-pyrrol-1-yl)carbamoyl moieties. The signals at 10.93 and 10.96 ppm for $4CH_3$, an intense resonance line at 103.11 ppm for 2CH–CH and the spectral lines at 165.11, 171.74 and 172.26 ppm due to the existing 4C=O groups in the ^{13}C NMR spectrum of molecule **18** proved the formation of the target bispyrrole derivative **18**.

BIOLOGY

The synthesised 5-oxopyrrolidine derivatives were investigated for the antibacterial activity against the gram-negative *Escherichia coli* and the gram-positive *Bacillus subtilis* bacteria strains. Inhibition zone tests were applied for the antibacterial evaluation. Generally, this assay is performed by the procedure [31] approved and published by the Clinical and Laboratory Standards Institute. ciprofloxacin was used as a control antibiotic, with an inhibition zone of 30 mm for both strains.

From the results of Table 2, bishydrazone **14b** bearing two 5-nitrothien-2-yl moieties with an inhibition zone of 19 mm for *E. coli* and 18 mm for *B. subtilis* was found to exhibit the strongest antibacterial activity among all the synthesised compounds. Monohydrazone **6a** with the same 5-nitrothien-2-yl moiety showed a slightly weaker inhibitory effect showing 18- and 17-mm inhibition zones, respectively. Among the synthesised azoles, the oxadiazolethione derivative **8** was the most potent exhibiting an inhibition zone of 10 mm against the gram-positive *B. subtilis*. All other compounds from the synthesised set were found to be inactive against the *E. coli* and *B. subtilis* bacteria strains.

CONCLUSIONS

In this study, we have presented novel 5-oxopyrrolidine derivatives that can act as antibacterial agents. For the synthesis of target compounds, acid hydrazide and asymmetric bis hydrazide were applied. The selected reaction methodologies were suitable for obtaining the intended hydrazones and azoles as well as asymmetric bis hydrazones and bis azoles in good yields.

Methyl 4-(4-(2-((5-nitrothien-2-yl)methylene)hydrazine-1-carbonyl)-2-oxopyrrolidin-1-yl)benzoate, *N*'-((5-nitrothien-2-yl)methylene) 1-(4-(2-((5-nitrothien-2-yl)methylene)hydrazine-1-carbonyl)phenyl)-5-oxopyrrolidine-3-carbohydrazide and 4-(2-oxo-4-(5-thioxo-4,5-dihydro-1,3,4-oxadiazol-2-yl)pyrrolidin-1-yl) benzoic acid showed a promising activity against the gram-positive *Bacillus subtilis* and the gram-negative *Escherichia coli* bacteria strains in comparison to ciprofloxacin.

The asymmetric bishydrazone bearing two 5-nitrothienyl cores in the molecule demonstrated the best activity against both bacteria tested. To disclose the antibacterial properties and the exact mode of action of these compounds detailed studies are required.

Received 21 September 2023
Accepted 10 October 2023

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1-(4-PAKEISTŲJŲ FENIL)-5-OKSOPIROLIDIN-3-KARBOHIDRAZIDO DARINIŲ SINTEZĖ IR PIRMINIAI ANTIBAKTERINIAI TYRIMAI

S a n t r a u k a

Šiame darbe įvykdyta įvairių metil 4-(4-(hidrazinokarbonil)-2-oksopirolidin-1-il)benzoato ir 1-(4-(hidrazinokarbonil)fenil)-5-oksopirolidin-3-karbohidrazido darinių sintezė. Susintetinta daug naujų mono- ir bishidrazonų bei azolų, į pastarųjų struktūrą įtraukiant 2,5-dimetilpirolo ir 3,5-dimetilpirazolo ir 1,3,4-oksadiazoltiono fragmentus. Hidrazonams gauti naudota įprasta hidrazidų kondensacijos su aromatiniais aldehidais metodika. Plečiant darinių įvairovę buvo atlikta mono- ir dihidrazidų kondensacija su izatinu, kurios metu atitinkamai gauti metil 4-(2-okso-4-(2-(2-oksoindolin-3-iliden)hidrazino-1-karbonil) pirolidin-1-il)benzoatas ir 5-okso-*N*'-(2-oksoindolin-3-iliden)-1-(4-(2-(2-oksoindolin-3-iliden)hidrazino-1-karbonil)fenil)pirolidin-3-karbohidrazidas, savo molekulių struktūrose turintys vieną arba du oksoindolin-3-ilideno fragmentus. Pirazolo ir pirolo dariniai gauti rūgšties dihidrazidui reaguojant atitinkamai su diketonais pentan-2,4-dionu ir heksan-2,5-dionu. Vieną oksadiazoltiono fragmentą turintis junginys buvo susintetintas vykdant monokarbohidrazido reakciją su anglies disulfidu šarminiame metanolio tirpale. Reakcija vyko per tarpinį ditiokarbazato druskos darinį, kurios vandeninį tirpalą parūgštinus koncentruota druskos rūgštimi buvo gautas ciklinis produktas – 4-(2-okso-4-(5-tiokso-4,5-dihidro-1,3,4-oksadiazol-2-il)pirolidin-1-il)benzoinė rūgštis. Reikia paminėti, kad buvo išskirta benzoinė rūgštis, bet ne metil benzoatas, kaip buvo tikėtasi. Manoma, kad dėl koncentruotos druskos rūgšties vyko esterinės grupės rūgštinė hidrolizė, todėl ir buvo gauta atitinkama karboksirūgštis.

Vykdant hidrazido reakciją su dietilmalonatu ledinėje acto rūgštyje tikėtasi gauti ciklinį metil 4-(4-(3,5-dioksopirazolidin-1-karbonil)-2-oksopirolidin-1-il)benzoatą, tačiau akivaizdu, kad reakcijos metu dėl acto rūgšties vyko visiškas hidrazido grupės acetilinimas ir iš reakcijos mišinio buvo išskirtas linijinės struktūros darinys – metil 4-(4-(2-acetilhidrazino-1-karbonil)-2-oksopirolidin-1-il)benzoatas.

Visi susintetinti junginiai identifikuoti remiantis ^{1}H, ^{13}C BMR, IR spektroskopijos ir elementinės analizės duomenimis. Pasirinktų reakcijos metodų patrauklumas – paprastos metodikos, tikslinių junginių išskyrimas bei išgryninimas, lemiantis geras gautų produktų išeigas, – daugeliu atvejų įrodo jų tinkamumą šios klasės junginių sintezei.

Taip pat darbe buvo atlikti pirminiai antibakteriniai susintetintų 5-oksopirolidino darinių tyrimai prieš gram-teigiamą *Bacillus subtilis* ir gram-neigiamą *Escherichia coli* bakterijų padermes. Tyrimai atlikti difuzijos į agarą metodu, įvertinant bakterijų slopinimo zoną (mm). Kontroliniu antibiotiku naudotas ciprofloksacinas, kurio abiejų bakterijų slopinimo zona yra 30 mm. Antibakterinis įvertinimas atskleidė, kad du 5-nitro-2-tienil fragmentus turintis asimetrinis bishidrazonas **14b** stipriausiai veikė abi tirtas bakterijų padermes: *E. coli* slopinimo zona buvo 19 mm, o *B. subtilis* – 18 mm. Labai panašiai šiuos patogenus veikė ir vieną 5-nitro-2-tienil fragmentą turintis hidrazonas **6a**. Dėl jo *E. coli* slopinimo zona buvo 18 mm, o *B. subtilis* – 17 mm. Iš tirtų azolo darinių vienintelis 1,3,4-oksadiazoltionas pasižymėjo slopinamuoju poveikiu. Jis selektyviai veikė *B. subtilis*, slopindamas jas tris kartus silpniau negu naudotas kontrolinis antibiotikas.

Biologinių tyrimų rezultatai rodo, kad 5-oksopirolidino dariniai, suformuoti monohidrazido **1a** ir dihidrazido **1b** pagrindu, gali būti toliau tiriami ieškant ir kuriant tikslinius, prieš gram-teigiamas ir gram-neigiamas bakterijų padermes nukreiptus antibakterinius preparatus.