CHEMIJA
2023. Vol. 34. No. 1. P. 57–69

ISSN 0235-7216 eISSN 2424-4538
DOI: https://doi.org/10.6001/chemija.2023.34.1.7

Synthesis, transformation and preliminary bioassay of 3-(thiazol-2-yl(*p*-tolyl)amino)propanoic acid derivatives

Birutė Sapijanskaitė-Banevič[1],

Birutė Grybaitė[1],

Rita Vaickelionienė[1*],

Ingrida Bružaitė[2]

[1] *Kaunas University of Technology,*
19 Radvilėnų Road,
50254 Kaunas, Lithuania

[2] *Vilnius Gediminas Technical University,*
11 Saulėtekio Avenue,
10223 Vilnius, Lithuania

1,3-Thiazole is one of the most attractive cores in heterocycles chemistry and drug discovery. Its widespread use in diverse medicinal substances makes thiazole a versatile scaffold, which leads to the novel generation of efficient pharmaceuticals. This study was intended to synthesize a series of *N,N*-disubstituted aminothiazole derivatives having various fragments at the 4th and 5th positions of the thiazole ring. The formation of such derivatives was carried out via the Hantsch reaction, condensation reactions with various aldehydes, bromination and formation of the chalcone-type derivatives. Another goal of this study was to investigate the antibacterial properties of the obtained compounds against some gram-positive *Bacillus* species such as *B. coagulans*, *B. subtilis* and *B. megaterium* and gram-negative *Escherichia coli*. As it is shown by our studies in recent years, the synthesized thiazoles can serve for future development of potent thiazole derivatives for various medicinal targets.

Keywords: thiazole, β-amino acids, chalcone, antibacterial activity

INTRODUCTION

Despite a large progress in medicine with a broad spectrum of antibiotics and antimicrobial agents, the incidence of life-threatening diseases caused by multi-drug pathogenic microorganisms has increased worldwide and is a major cause of morbidity and mortality in immunocompromised patients [1]. To control this prevalence and reduce antimicrobial resistance, more effective new antibacterial drugs without side effects are needed.

The thiazole scaffold is a significant heterocyclic structure present in a large variety of biologically active compounds, and it is also a unique essential scaffold of many natural often marine-derived sources [2]. Thiazole-based synthetic compounds are a versatile building block for drug discovery and design. At last decades, thiazole derivatives have focused a considerable attention because of their broad application including pharmaceutical and biological activities [3]. The thiazole motif appears in the structure of penicillin and bacitracin antibiotics [4] and a wide variety of synthetic drugs such as sulfathiazole and acinitrazole [5] used as antimicrobial agents, antidepressant pramipexole with antiparkinsonian activity, anti-inflammatory drug meloxicam, antineoplastic agent tiazofurin [6, 7], antiasthmatic drug cinalukast [8], non-steroidal immunomodulator fanetizole [9], antiprotozoal drug nitazoxanide [10], antifungal agent abafungin, respiratory stimulant amiphenazole and anti-ulcer agent nizatidine [11].

In addition to the above-mentioned, our previous extensive studies in the field of functionalized 2-aminothiazoles showed that derivatives containing thiazole frameworks have a promising or even excellent

* Corresponding author. Email: rita.vaickelioniene@ktu.lt

antimicrobial activity [12–15], effectively promote the growth of agricultural crops such as rapeseed, increase their seed yield and oil content [16], also strongly inhibit human carbonic anhydrases [17]. Such a variety of the properties influence the synthesis of thiazole-based compounds by modifying them by potential pharmacophoric groups at different positions with the aim to generate new structures and test their potential antimicrobial activity.

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 synthesized 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 B-540 melting point analyser (Büchi Corporation, New Castle, DE, USA) and were uncorrected. NMR spectra were recorded on a Brucker Avance III (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 Perkin–Elmer Spectrum BX FT–IR spectrometer using KBr pellets. Mass spectra were obtained on a Bruker maXis UHRTOF mass spectrometer with ESI ionization. Elemental analyses (C, H, N) were conducted using the Elemental Analyzer CE-440; their results were found to be in a good agreement (±0.3%) with the calculated values.

General procedure for the preparation of compounds 2a–c

A mixture of the corresponding thioureido acid **1a–c** (8 mmol), monochloroacetic acid (0.85 g, 9 mmol), sodium carbonate (1.06 g, 10 mmol) or sodium acetate (0.82 g, 10 mmol) in water (150 mL) was refluxed for 2–3 h, then cooled down and neutralized with diluted acetic acid to pH 6. The obtained precipitate was filtered off, washed with water, dried and recrystallized to afford the corresponding thiazolones **2a–c** as follows: **2a** – resynthesized according to the method described in Ref. [18], m.p.: 198–199°C; **2b –** a white solid (1.16 g, 44%, from water), m.p.: 197–198°C (from water); **2c –** a white solid (0.74 g, 28%, from water: 1,4-dioxane mixture, 1:1), m.p.: 195–196°C.

2-Methyl-3-((4-oxo-4,5-dihydrothiazol-2-yl)(*p*-tolyl)amino)propanoic acid (2b)

^{1}H NMR (300 MHz, $(CD_3)_2SO$, 25°C): δ = 12.45 (bs, 1H; OH), 7.33 (s, 4H; H_{ar}), 4.04–4.19 (m, 2H; NCH_2), 3.91 (s, 2H; SCH_2), 2.55–2.70 (m, 2H; CHCO), 2.36 (s, 3H; CH_3), 1.07 (d, *J* = 7.0 Hz, 3H; $CHCH_3$) ppm; ^{13}C NMR (75.4 MHz, $(CD_3)_2SO$, 25°C): δ = 186.94, 184.09, 175.05, 139.25, 137.78, 130.31, 127.68, 56.35, 40.53, 37.49, 20.76, 14.72 ppm; IR (KBr): ν = 1716, 1652 (CO), 1517 (C=N) cm^{-1}. Elemental analysis calcd (%) for $C_{14}H_{16}N_2O_3S$: C 57.52, H 5.52, N 9.58; found: C 57.65, H 5.49, N 9.78.

3-((4-Oxo-4,5-dihydrothiazol-2-yl)(*p*-tolyl)amino)butanoic acid (2c)

^{1}H NMR (300 MHz, $(CD_3)_2SO$, 25°C): δ = 12.45 (s, 1H; OH), 7.35 (d, *J* = 8.1 Hz, 2H; H_{ar}), 7.26 (d, *J* = 8.2 Hz, 2H; H_{ar}), 5.16 (h, *J* = 7.0 Hz, 1H; NCH), 3.84 (s, 2H; SCH_2), 2.57 (dd, *J* = 16.1, 7.2 Hz, 1H; CH_2CO), 2.37 (s, 3H; CH_3), 2.32 (dd, *J* = 16.1, 7.4 Hz, 1H; CH_2CO), 1.19 (d, *J* = 6.8 Hz, 3H; $CHCH_3$) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 186.98, 183.58, 171.77, 139.84, 134.39, 130.19, 129.59, 54.45, 39.87, 38.68, 20.79, 18.57 ppm; IR (KBr): ν = 1720, 1662 (CO), 1521 (C=N) cm^{-1}. Elemental analysis calcd (%) for $C_{14}H_{16}N_2O_3S$: C 57.52, H 5.52, N 9.58; found: C 57.43, H 5.56, N 9.53.

General procedure for the preparation of compounds 3–10

A mixture of thiazolone **2a** (1.8 mmol), the corresponding benzaldehyde or thiophenecarboxaldehyde (1.98 mmol), sodium carbonate (9.45 mmol) and water (10 mL) was heated at reflux for 3 h, then cooled down and acidified with 10% acetic acid to pH 6. The formed solid was filtered of, washed with water and recrystallized by dissolving it in 5% sodium carbonate (0.5 g/10 mL of water), filtering the solution and acidifying the filtrate with 10% acetic acid to pH 6. The solid was washed with water and dried to afford compounds **3–11** as follows: **3** – a light-yellow solid (0.22 g, 34% yield), m.p.: 184–185°C; **4** – a white solid (0.31 g, 46%

yield), m.p.: 219–220°C; **5** – a light-yellow solid (0.26 g, 36% yield), m.p.: 214–215°C; **6** – a light-brown solid (0.4 g, 50% yield), m.p.: 216–217°C; **7** – a white solid (0.24 g, 35% yield), m.p.: 227–228°C; **8** – an orange solid (0.25 g, 34% yield), m.p.: 137–138°C; **9** – a brown solid (0.43 g, 58% yield), m.p.: 237–238°C; **10** – a light-yellow solid (0.28 g, 46% yield), m.p.: 213–214°C.

3-((5-Benzylidene-4-oxo-4,5-dihydrothiazol-2-yl)(*p*-tolyl)amino)propanoic acid (3)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.45 (s, 1H; OH), 7.64 (s, 1H; C=CH), 7.46–7.35 (m, 9H; H_{ar}), 4.26 (t, *J* = 7.3 Hz, 2H; NCH_2), 2.62 (t, *J* = 7.3 Hz, 2H; CH_2CO), 2.40 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 179.51, 176.34, 171.90, 139.75, 137.31, 133.68, 133.06, 131.74, 130.56, 130.39, 130.02, 129.84, 129.44, 129.33, 129.23, 129.17, 127.93, 49.94, 31.84, 20.84 ppm; IR (KBr): ν = 1531, 1706, 1734, 3380 cm^{-1}. Elemental analysis calcd (%) for $C_{20}H_{18}N_2O_3S$: C 65.55, H 4.95, N 7.65; found: C 65.66, H 4.88, N 7.55.

3-((5-(4-Fluorobenzylidene)-4-oxo-4,5-dihydrothiazol-2-yl)(*p*-tolyl)amino)propanoic acid (4)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.53 (bs, 1H; OH), 7.65 (s, 1H; SC=CH), 7.53–7.48 (m, 2H; H_{ar}), 7.44 (d, *J* = 8.2 Hz, 2H; H_{ar}), 7.38 (d, *J* = 8.2 Hz, 2H; H_{ar}), 7.31–7.26 (m, 2H; H_{ar}), 4.25 (t, *J* = 7.4 Hz, 2H; NCH_2), 2.60 (t, *J* = 7.4 Hz, 2H; CH_2CO), 2.39 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 179.44, 176.16, 172.02, 163.74, 161.26, 139.72, 137.31, 131.82, 131.73, 130.55, 130.35, 130.32, 129.25, 128.94, 128.92, 127.91, 116.45, 116.23, 50.07, 31.99, 20.82 ppm; IR (KBr): ν = 1521, 1713, 3416 cm^{-1}. Elemental analysis calcd (%) for $C_{20}H_{17}FN_2O_3S$: C 62.49, H 4.46, N 7.29; found: C 62.26, H 4.50, N 7.18.

3-((5-(4-Chlorobenzylidene)-4-oxo-4,5-dihydrothiazol-2-yl)(*p*-tolyl)amino)propanoic acid (5)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.45 (bs, 1H; OH), 7.63 (s, 1H; SC=CH), 7.50 (d, *J* = 8.2 Hz, 2H; H_{ar}), 7.47–7.42 (m, 4H; H_{ar}), 7.38 (d, *J* = 8.2 Hz, 2H; H_{ar}), 4.26 (t, *J* = 7.3 Hz, 2H; NCH_2), 2.61 (t, *J* = 7.3 Hz, 2H; CH_2CO), 2.40 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 179.38, 176.11, 171.88, 139.78, 137.26, 134.33, 132.60, 131.08, 130.59, 129.89, 129.31, 129.07, 127.92, 50.03, 31.82, 20.84 ppm; IR (KBr): ν = 1518, 1715, 3410 cm^{-1}. Elemental analysis calcd (%) for $C_{20}H_{17}ClN_2O_3S$: C 59.92, H 4.27, N 6.99; found: C 59.98, H 4.18, N 6.87.

3-((5-(4-Bromobenzylidene)-4-oxo-4,5-dihydrothiazol-2-yl)(*p*-tolyl)amino)propanoic acid (6)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 7.64–7.36 (m, 9H; H_{ar}, SC=CH), 4.23 (t, *J* = 7.6 Hz, 2H; NCH_2), 2.55 (t, *J* = 7.5 Hz, 2H; CH_2CO), 2.39 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 179.38, 175.92, 172.16, 139.69, 137.32, 132.95, 132.20, 131.22, 130.54, 130.04, 129.01, 127.90, 123.12, 50.49, 32.42, 20.82 ppm; IR (KBr): ν = 1511, 1717, 3390 cm^{-1}. Elemental analysis calcd (%) for $C_{20}H_{17}BrN_2O_3S$: C 53.94, H 3.85, N 6.29; found: C 53.78, H 3.77, N 6.21.

3-((5-(4-Methylbenzylidene)-4-oxo-4,5-dihydrothiazol-2-yl)(*p*-tolyl)amino)propanoic acid (7)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 7.59 (s, 1H; SC=CH), 7.43–7.22 (m, 8H; H_{ar}), 4.21 (t, *J* = 7.6 Hz, 2H; NCH_2), 2.51 (2H; CH_2CO, overlaps with the signal of the DMSO-d_6), 2.38 (s, 3H; CH_3), 2.28 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 179.64, 176.02, 172.34, 139.85, 139.59, 137.46, 130.95, 130.50, 130.27, 129.80, 129.43, 128.14, 127.92, 50.60, 32.80, 21.02, 20.81 ppm; IR (KBr): ν = 1511, 1733, 3419 cm^{-1}. Elemental analysis calcd (%) for $C_{21}H_{20}N_2O_3S$: C 66.30, H 5.30, N 7.36; found: C 66.23, H 5.21, N 7.27.

3-((5-(4-(Dimethylamino)benzylidene)-4-oxo-4,5-dihydrothiazol-2-yl)(*p*-tolyl)amino) propanoic acid (8)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 7.51 (s, 1H; SC=CH), 7.42 (d, *J* = 8.1 Hz, 2H; H_{ar}), 7.37 (d, *J* = 8.1 Hz, 2H; H_{ar}), 7.25 (d, *J* = 8.6 Hz, 2H; H_{ar}), 6.72 (d, *J* = 8.6 Hz, 2H; H_{ar}), 4.22 (t, *J* = 7.5 Hz, 2H; NCH_2), 2.98, 2.93 (2S, 6H; $N(CH_3)_2$), 2.59 (t, *J* = 7.5 Hz, 2H; CH_2CO), 2.39 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 180.11, 175.88, 172.21, 150.07, 139.48, 137.67, 131.60, 131.45, 131.29, 130.50, 128.03, 122.42, 120.53, 112.04, 49.74, 32.15, 20.83 ppm. Elemental analysis calcd (%) for $C_{22}H_{23}N_3O_3S$: C 64.53, H 5.66, N 10.26; found: C 64.41, H 5.50, N 10.11.

3-((5-(4-Nitrobenzylidene)-4-oxo-4,5-dihydrothiazol-2-yl)(*p*-tolyl)amino)propanoic acid (9)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 7.59 (s, 1H; SC=CH), 7.43–7.22 (m, 8H; H_{ar}), 4.24 (t, *J* = 7.6 Hz, 2H; NCH_2), 2.56 (t, *J* = 7.5 Hz, 2H; CH_2CO), 2.37 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 179.91, 175.89, 172.22, 159.41, 139.51, 137.63, 131.47, 130.56, 130.37, 127.87, 124.61, 124.27, 116.17, 115.81, 50.21, 32.51, 20.81 ppm. IR (KBr): ν = 1517, 1721 and 3413 cm^{-1}. Elemental analysis calcd (%) for $C_{20}H_{17}N_3O_5S$: C 58.39, H 4.16, N 10.21; found: C 58.39, H 4.11, N 10.17.

3-((4-Oxo-5-(thiophen-2-ylmethylene)-4,5-dihydrothiazol-2-yl)(*p*-tolyl)amino)propanoic acid (10)

A mixture of thiazolone **2a** (0.5 g, 1.8 mmol), thiophenecarboxaldehyde (0.22 g, 1.98 mmol), sodium carbonate (1 g, 9.45 mmol) and water (10 mL) was heated at reflux for 3 h, then cooled down and acidified with 10% acetic acid to pH 6. The formed solid was filtered off, washed with water and recrystallized by dissolving it in 5% sodium carbonate (0.5 g/10 mL of water), filtering the solution and acidifying the filtrate with 10% acetic acid to pH 6. The solid was washed with water and dried to afford compound **10** as a light-yellow solid (0.31 g, 46% yield), m.p.: 213–214°C.

^{1}H NMR (700 MHz, $(CD_3)_2SO$, 25°C): δ = 12.26 (s, 1H; OH), 7.85 (s, 1H; SC=CH), 7.77 (d, *J* = 5.0 Hz, 1H; H_{th}), 7.51 (d, *J* = 5.0 Hz, 1H; H_{th}), 7.44 (d, *J* = 8.1 Hz, 2H; H_{ar}), 7.38 (d, *J* = 8.1 Hz, 2H; H_{ar}), 7.18 (dd, *J* = 5,0, 3.7 Hz, 1H; H_{th}), 4.22 (t, *J* = 7.5 Hz, 2H; NCH_2), 2.56 (t, *J* = 7.5 Hz, 2H; CH_2CO), 2.40 (s, 3H; CH_3) ppm; ^{13}C NMR (176 MHz, $(CD_3)_2SO$, 25°C): δ = 179.23, 175.28, 172.12, 139.70, 138.42, 137.43, 133.24, 131.32, 130.50, 128.81, 127.95, 127.39, 123.23, 50.20, 32.35, 20.82 ppm; IR (KBr): ν = 1519, 1705, 3417 cm^{-1}. Elemental analysis calcd (%) for $C_{18}H_{16}N_2O_3S$: C 58.05, H 4.33, N 7.52; found: C 58.22, H 4.28, N 7.38.

3,3'-((1,4-Phenylenebis(methaneylylidene)) bis(4-oxo-4,5-dihydrothiazole-2-yl-5-ylidene)) bis(*p*-tolylazanediyl))dipropanoic acid (11)

A mixture of thiazolone **2a** (0.5 g, 1.8 mmol), terephthalaldehyde (0.48 g, 3.6 mmol), sodium carbonate (1.01 g, 9.5 mmol) and water (10 mL) was heated at reflux for 3 h, then cooled down and acidified with 10% acetic acid to pH 6. The formed solid was filtered off, washed with water and recrystallized by dissolving it in 5% sodium carbonate (0.5 g/10 mL of water), filtering the solution and acidifying the filtrate with 10% acetic acid to pH 6. The solid was washed with water and dried to afford compound **11** as a yellow solid (0.45 g, 38% yield), m.p.: 268–269°C.

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 7.60 (s, 2H; 2SC=CH), 7.51 (s, 4H; H_{ar}), 7.44 (d, *J* = 8.2 Hz, 4H; H_{ar}), 7.38 (d, *J* = 8.2 Hz, 4H; H_{ar}), 4.25 (t, *J* = 7.3 Hz, 4H; $2NCH_2$), 2.60 (t, *J* = 7.3 Hz, 4H; $2CH_2CO$), 2.41 (s, 6H; $2CH_3$) ppm; IR (KBr): ν = 1511, 1716, 3419 cm^{-1}. Elemental analysis calcd (%) for $C_{34}H_{30}N_4O_6S_2$: C 62.37, H 4.62, N 8.56; found: C 62.27, H 4.54, N 8.49.

3-((5,5-Dibromo-4-oxo-4,5-dihydrothiazol-2-yl)(*p*-tolyl)amino)propanoic acid (12)

To a mixture of thiazolone **2a** (1 g, 3.59 mmol) and glacial acetic acid (5 mL), a solution of bromine (2.01 g, 0.644 mL, 12.59 mmol) in acetic acid (3 mL) was added dropwise and the mixture was stirred at 60°C for 4 h. Then the mixture was cooled down and diluted with aqueous sodium acetate solution (1 g/50 mL of water) with the subsequent addition of aqueous $Na_2S_2O_3$ solution to neutralize unreacted bromine. The formed solid was filtered off, washed with water, dried and recrystallized from 1,4-dioxane to afford compound **12** as a light-yellow solid (0.39 g, 25% yield), m.p.: 300°C (decomp.).

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.44 (s, 1H; OH), 7.49–7.34 (m, 4H; H_{ar}), 4.23 (t, *J* = 7.0 Hz, 2H; NCH_2), 2.59 (t, *J* = 7.2 Hz, 2H; CH_2CO), 2.41 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 179.79, 179.06, 171.84, 140.12, 136.98, 130.60, 129.09, 127.84, 49.97, 31.78, 20.85 ppm; IR (KBr): ν = 1511, 1683, 1725, 3441 cm^{-1}. Elemental analysis calcd (%) for $C_{13}H_{12}Br_2N_2O_3S$: C 35.80, H 2.77, N 6.42; found: C 35.75, H 2.72, N 6.38.

3-(1,3-Thiazol-2-yl(*p*-tolyl)amino)propanoic acid (13)

A mixture of thioureido acid **1a** (1.5 g, 6.29 mmol), chloroacetaldehyde 50% aqueous solution (0.56 mL, 8.8 mmol) and dry acetone (15 mL) was refluxed for 3 h. Then acetone was distilled under reduced pressure and the reaction mixture was diluted with water (40 mL) and sodium acetate (0.82 g, 10 mmol) was added, and the mixture was stirred for 1 h at room temperature. The formed precipitate was filtered off

and washed with water and recrystallized form ethanol to afford compound **13** as a very light-yellow solid (1.29 g, 78% yield), m.p.: 141–142°C.
^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.24 (s, 1H; OH), 7.28 (s, 4H; H_{ar}), 7.17 (d, *J* = 3.6 Hz, 1H; CH_{th}), 6.68 (d, *J* = 3.6 Hz, 1H; CH_{th}), 4.07 (t, *J* = 7.3 Hz, 2H; NCH_2), 2.59 (t, *J* = 7.3 Hz, 2H; CH_2CO), 2.33 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 172.50, 169.95, 142.31, 139.13, 136.82, 130.52, 129.50, 126.74, 107.94, 48.44, 32.30, 20.60 ppm; IR (KBr): ν = 1518, 1683, 1718, 3431 cm^{-1}. Elemental analysis calcd (%) for $C_{13}H_{14}N_2O_2S$: C 59.52, H 5.38, N 10.68; found: C 59.44, H 5.29, N 10.57.

14, 16 and **17** were resynthesized according to the methods described in [19].

3-((4-(3,4-Dichlorophenyl)thiazol-2-yl)(*p*-tolyl)amino)propanoic acid (15)

A mixture of thioureido acid **1a** (1.19 g, 5 mmol), 2-bromo-3,4-dichloroacetophenone (2.21 g, 6 mmol) and sodium acetate (0.57 g, 7 mmol) was refluxed in propan-2-ol (10 mL) for 4 h. After completion of the reaction the reaction mixture was diluted with water (40 mL), and the formed precipitate was purified by dissolving it in 15% aqueous sodium carbonate, filtering and acidifying the filtrate with acetic acid to pH 6 to afford compound **15** as a light-greyish solid (1.47 g, 72% yield), m.p.: 161–162°C.
^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.28 (s, 1H; OH), 8.08 (d, *J* = 2.0 Hz, 1H; H_{ar}), 7.86, 7.84 (2d, *J* = 2.1 Hz, 1H; H_{ar}), 7.66, 7.64 (2s, 1H; H_{ar}), 7.34–7.28 (m, 5H; H_{ar}, S-CH), 4.17 (t, *J* = 7.2 Hz, 2H; NCH_2), 2.65 (t, *J* = 7.2 Hz, 2H; CH_2CO), 2.35 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 172.50, 169.41, 147.71, 141.70, 137.22, 135.18, 131.32, 130.73, 130.59, 129.61, 127.12, 126.93, 125.66, 104.78, 48.47, 32.31, 20.62 ppm. IR (KBr): ν = 1517, 1721, 3413 cm^{-1}. Elemental analysis calcd (%) for $C_{19}H_{16}Cl_2N_2O_2S$: C 56.03, H 3.96, N 6.88; found: C 56.22, H 4.02, N 6.75.

3-((4-(Naphthalen-2-yl)thiazol-2-yl)(*p*-tolyl)amino)propanoic acid (18)

A mixture of thioureido acid **1a** (0.16 g, 0.68 mmol) and 2-bromo-2-acetonaphthone (0.2 g, 0.81 mmol) was refluxed for 4 h in dry acetone (10 mL). Then a solution of sodium acetate (0.13 g, 1.6 mmol) in water (10 mL) was added and the mixture was stirred for 1 h. The formed precipitate was filtered off, washed with water and recrystallized from methanol to afford compound **18** as a light grey solid (0.16 g, 60% yield), m.p.: 176–177°C.
^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.30 (s, 1H; OH), 8.41 (s, 1H; H_{ar}), 8.02, 8.00 (2d, *J* = 1.7 Hz, 1H; H_{ar}), 7.96–7.87 (m, 3H; H_{ar}), 7.54–7.45 (m, 2H; H_{ar}), 7.38–7.28 (m, 4H; H_{ar}), 7.27 (s, 1H; SCH), 4.25 (t, *J* = 7.2 Hz, 2H; NCH_2), 2.71 (t, *J* = 7.2 Hz, 2H; CH_2CO), 2.36 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 172.60, 169.31, 150.31, 141.89, 137.09, 133.12, 132.43, 132.13, 130.57, 128.07, 127.97, 127.53, 126.97, 126.34, 125.87, 124.07, 103.44, 48.41, 32.43, 20.64 ppm. Elemental analysis calcd (%) for $C_{23}H_{20}N_2O_2S$: C 71.11, H 5.19, N 7.21; found: C 71.17, H 5.10, N 7.28.

3-((4-(2-Oxo-2*H*-chromen-3-yl)thiazol-2-yl)(*p*-tolyl)amino)propanoic acid (19)

A mixture of thioureido acid **1a** (0.13 g, 0.55 mmol) and 3-(bromoacetyl)-2-acetonaphthone (0.18 g, 0.66 mmol) was refluxed for 4 h in dry acetone (10 mL). Afterwards, the reaction mixture was cooled down and diluted with water (25 mL). Then sodium acetate (0.13 g, 1.5 mmol) was added and the mixture was stirred for 20 min. The formed precipitate was filtered off and washed with water and diethyl ether, dried and recrystallized from methanol to afford compound **19** as a white solid (0.118 g, 54% yield), m.p.: 115–116°C.
^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.28 (s, 1H; OH), 8.64 (d, *J* = 3.4 Hz, 1H; H_{ar}), 7.90–7.84 (m, 1H; H_{ar}), 7.64–7.59 (m, 1H; H_{ar}), 7.57 (s, 1H; H_{ar}), 7.43 (d, *J* = 8.3 Hz, 1H; H_{ar}), 7.41–7.25 (m, 5H; $H_{ar,}$ S–CH), 4.32–4.14 (m, 2H; NCH_2), 2.76 (t, *J* = 7.1 Hz, 1H; CH_2CO), 2.67 (t, *J* = 7.1 Hz, 1H; CH_2CO), 2.35 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 172.65, 171.60, 168.66, 168.57, 158.74, 158.72, 152.28, 143.80, 143.79, 141.71, 141.65, 138.43, 137.42, 137.36, 131.59, 131.57, 130.65, 128.80, 128.74, 127.09, 127.05, 124.69, 120.40, 119.26, 115.86, 115.84, 109.71, 109.64, 48.31, 48.22, 32.39, 32.26, 20.66 ppm. IR (KBr): ν = 1517, 1721, 3413 cm^{-1}. Elemental analysis calcd (%) for $C_{22}H_{18}N_2O_4S$: C 65.01, H 4.46, N 6.89; found: C 65.10, H 4.48, N 6.82.

3-((4,9-Dioxo-4,9-dihydronaphtho[2,3-d]thiazol-2-yl)(*p*-tolyl)amino)propanoic acid (20)

A mixture of thioureido acid **1a** (2 g, 8.3 mmol), 2,3-dichloronaphthalene-1,4-dione (2.28 g, 12.6 mmol) and sodium acetate (1.2 g, 14.6 mmol)

in dry acetone (30 mL) was refluxed for 4 h. Then the solvent was evaporated at the reduced pressure, the obtained precipitate was dissolved in aqueous 5% potassium hydroxide (100 mL), filtered off, and the filtrate was neutralized with glacial acetic acid. The obtained solid was recrystallized from propan-2-ol to afford compound **20** as a brown solid (1 g, 30% yield), m.p.: 163–164°C. ^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.39 (s, 1H; OH), 8.08–7.89 (m, 3H; H_{ar}), 7.77 (t, *J* = 7.6 Hz, 1H; H_{ar}), 7.58 (t, *J* = 7.6 Hz, 1H; H_{ar}), 7.46–7.37 (m, 3H; H_{ar}), 4.34 (t, *J* = 7.2 Hz, 2H; NCH_2), 2.69 (t, *J* = 7.2 Hz, 2H; CH_2CO), 2.40 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 180.01, 175.89, 175.62, 172.13, 169.99, 160.28, 142.40, 140.12, 139.08, 135.13, 134.63, 131.87, 130.99, 130.80, 128.84, 127.05, 126.84, 125.85, 120.71, 49.13, 32.16, 20.78 ppm; IR (KBr): ν =1534, 1685, 1721, 3410 cm^{-1}. Elemental analysis calcd (%) for $C_{21}H_{16}N_2O_4S$: C 64.27, H 4.11, N 7.14; found: C 64.22, H 4.18, N 7.38.

General procedure for the preparation of thiazoles 21a–c and 22a–c

A mixture of the corresponding thioureido acid **1a–c** (1.20 g, 5 mmol), chloroacetone or 2-bromoacetophenone (6 mmol) and dry acetone (15 mL) was refluxed for 4 h. Then the reaction mixture was diluted with water (30 mL), and sodium acetate (0.82 g, 10 mmol) was added, and the mixture was stirred for 10 min at room temperature. The formed precipitate was filtered off, washed with water and recrystallized from the appropriate solvent to afford compounds **21** and **22** as follows: **21b –** a grey solid (0.61 g, 44%), m.p.: 105–106°C; **21c** – a yellowish solid (0.63 g, 46%), m.p.: 137–138°C; **22b** – a yellow solid (0.91g, 54%), m.p.: 140–141°C; **22c** – a greenish solid (1 g, 60%), m.p.: 134–135°C.

3-((4-Methylthiazol-2-yl)(*p*-tolyl)amino)propanoic acid (21a) was resynthesized according to Ref. [20].

2-Methyl-3-((4-methylthiazol-2-yl)(*p*-tolyl) amino)propanoic acid (21b)

^{1}H NMR (400 MHz, $CDCl_3$, 25°C): δ = 12.72 (s, 1H; OH), 7.29 (d, *J* = 8.4 Hz, 2H; H_{ar}), 7.34 (d, *J* = 8.4 Hz, 2H; H_{ar}), 6.10 (s, 1H; S-CH), 4.50 (dd, *J* = 14.6, 5.5 Hz, 1H; NCH_2), 3.74 (dd, *J* = 14.5, 4.4 Hz, 1H; NCH_2), 3.15–3.02 (m, 1H; CH), 2.47 (s, 3H; CH_3), 2.37 (s, 3H; CH_3), 1.33 (d, *J* = 7.1 Hz, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $CDCl_3$, 25°C): δ = 176.93, 171.12, 146.97, 142.71, 138.57, 131.11, 126.67, 102.20, 57.78, 40.81, 21.27, 16.55, 16.27 ppm. IR (KBr): ν = 1506, 1715, 3459 cm^{-1}. Elemental analysis calcd (%) for $C_{15}H_{18}N_2O_2S$: C 62.04, H 6.25, N 9.65; found: C 62.10, H 6.28, N 9.68.

3-((4-Methylthiazol-2-yl)(*p*-tolyl)amino) butanoic acid (21c)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.29 (s, 1H; OH), 7.30 (d, *J* = 8.3 Hz, 2H; H_{ar}), 7.18 (d, *J* = 8.3 Hz, 2H; H_{ar}), 6.16 (s, 1H; S-CH), 4.98 (h, *J* = 6.8 Hz, 1H; NCH), 2.63 (dd, *J* = 15.6, 6.7 Hz, 1H; CH_2CO), 2.35 (s, 3H; CH_3), 2.32–2.23 (m, 1H; CH_2CO), 2.14 (s, 3H; CH_3), 1.18 (d, *J* = 6.7 Hz, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 172.35, 169.94, 148.22, 138.77, 138.08, 130.57, 130.10, 101.62, 52.09, 39.39, 20.73, 18.49, 17.56 ppm. IR (KBr): ν = 1505, 1715, 3460 cm^{-1}. Elemental analysis calcd (%) for $C_{15}H_{18}N_2O_2S$: C 62.04, H 6.25, N 9.65; found: C 61.98, H 6.21, N 9.69.

3-((4-Phenylthiazol-2-yl)(*p*-tolyl)amino)propanoic acid (22a) was resynthesized according to Ref. [20]. m.p.: 156–157°C.

2-Methyl-3-((4-phenylthiazol-2-yl)(*p*-tolyl) amino)propanoic acid (22b)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.29 (s, 1H; OH), 7.86 (d, *J* = 7.6 Hz, 2H; H_{ar}), 7.42–7.26 (m, 7H; H_{ar}), 7.11 (s, 1H; S-CH), 4.17 (dd, *J* = 13.7, 7.2 Hz, 1H; NCH_2), 4.07 (dd, *J* = 13.7, 7.2 Hz, 1H; NCH_2), 2.88 (h, *J* = 7.1 Hz, 1H; CH), 2.35 (s, 3H; CH_3), 1.14 (d, *J* = 7.0 Hz, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, (400 MHz, $(CD_3)_2SO$, 25°C): δ = 175.85, 169.83, 150.28, 142.22, 137.00, 134.68, 130.59, 128.55, 127.51, 126.89, 125.64, 102.53, 55.15, 38.04, 20.69, 14.98 ppm. IR (KBr): ν = 1511, 1714 cm^{-1}. Elemental analysis calcd (%) for $C_{20}H_{20}N_2O_2S$: C 68.16, H 5.72, N 7.95; found: C 68.21, H 5.75, N 8.05.

3-((4-Phenylthiazol-2-yl)(*p*-tolyl)amino) butanoic acid (22c)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.28 (s, 1H; OH), 7.86 (d, *J* = 7.5 Hz, 2H; H_{ar}), 7.45–7.22 (m, 7H; H_{ar}), 7.06 (s, 1H; S-CH), 5.07 (h, *J* = 7.0 Hz,

1H; NCH), 2.78 (dd, *J* = 15.6, 6.8 Hz, 1H; CH_2CO), 2.42 (dd, *J* = 15.6, 6.8 Hz, 1H; CH_2CO), 2.37 (s, 3H; CH_3), 1.28 (d, *J* = 6.7 Hz, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 172.45, 169.78, 150.29, 146.06, 139.13, 138.24, 134.84, 130.69, 129.84, 128.53, 127.44, 125.64, 102.33, 52.86, 39.31, 20.77, 18.46 ppm. IR (KBr): ν = 1513, 1712 cm^{-1}. Elemental analysis calcd (%) for $C_{20}H_{20}N_2O_2S$: C 68.16, H 5.72, N 7.95; found: C 68.22, H 5.77, N 8.05.

3-((5-Acetyl-4-methylthiazol-2-yl)(*p*-tolyl) amino)propanoic acid (23)

A mixture of thioureido acid **1a** (1.5 g, 0.0062 mol), 3-chloropentane-2,4-dione and acetone (20 mL) was refluxed for 4 h. Then the reaction mixture was diluted with water, and sodium acetate (1.46 g, 0.0178 mol) was added. And the mixture was stirred 15 min at room temperature. The formed precipitate was filtered off, washed with water and diethyl ether and recrystallized from methanol to give compound **23** as a white solid (1.50 g, 76% yield), m.p.: 175–176°C.

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.31 (s, 1H; OH), 7.32 (s, 4H; H_{ar}), 4.12 (t, *J* = 7.3 Hz, 2H; NCH_2), 2.57 (t, *J* = 7.3 Hz, 2H; CH_2CO), 2.49 (s, 3H; CH_3), 2.35 (s, 3H; CH_3), 2.29 (s, 3H; CH_3) ppm; ^{13}C NMR (100 MHz, $(CD_3)_2SO$, 25°C): δ = 188.54, 172.21, 170.51, 157.35, 140.71, 138.13, 130.77, 127.02, 122.29, 47.95, 32.20, 29.52, 20.66, 18.47. IR (KBr): ν = 3423, 1731, 1679, 1569 cm^{-1}. Elemental analysis calcd (%) for $C_{16}H_{18}N_2O_3S$: C 60.36, H 5.70, N 8.80; found: C 60.30, H 5.65, N 8.74.

General procedure for the preparation of compounds 24–26

After stirring a solution of compound **23** (0.5 g, 1.6 mmol) in 30% aqueous NaOH solution (16 mL) at 80°C temperature, the corresponding aromatic aldehyde (1.92 mmol) was added and stirred at 80°C temperature 2 h. Then the reaction mixture was cooled down, the precipitate was filtered off, washed with saturated aqueous sodium chloride solution and purified by dissolving it in 10 mL water, filtering and acidifying the filtrate with glacial acetic acid to pH 5. The obtained compounds **24–26** were recrystallized from propan-2-ol to afford yellow solids as follows: **24** – (0.4 g, 61%), m.p.: 135–136°C; **25** – (0.38 g, 56%), m.p.: 141–142°C; **26** – (0.34 g, 49%), m.p.: 133–134°C.

3-((5-Cinnamoyl-4-methylthiazol-2-yl)(*p*-tolyl) amino)propanoic acid (24)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.35 (br. s, 1H; OH), 7.74–7.70 (m, 2H; H_{ar}), 7.53 (d, 1H, *J* = 15.4 Hz; COCH=CH), 7.41–7.29 (m, 7H; H_{ar}), 7.20 (d, 1H, *J* = 15.4 Hz; COCH=CH), 4.16 (t, 4H, *J* = 7.4 Hz; NH_2), 2.61 (s, 3H; $NCCH_3$), 2.58 (t, 2H, *J* = 7.4 Hz; CH_2CO), 2.37 (s, 3H; CH_3). ^{13}C NMR (101 MHz, $(CD_3)_2SO$, 25°C): δ = 180.28, 172.28, 170.83, 158.77, 141.61, 140.66, 138.24, 134.48, 130.87, 130.30, 128.90, 128.48, 127.07, 124.63, 122.19, 48.18, 32.23, 20.74, 19.04. IR (KBr): ν = 1725, 1622, 1520 cm^{-1}. HRMS (ESI) for $C_{23}H_{22}N_2O_3S$, $[M+H]^+$: 407.1351, found: 407.1437 $[M+H]^+$.

3-((5-(3-(4-Fluorophenyl)acryloyl)-4-methylthiazol-2-yl)(*p*-tolyl)amino)propanoic acid (25)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.35 (br. s, 1H; OH), 7.80 (dd, 2H, *J* = 8.6; 5,5 Hz; H_{ar}), 7.53 (d, 1H, *J* = 15.4 Hz; COCH=CH), 7.35 (s, 4H; H_{ar}), 7,22 (t, 2H, *J* = 8,7 Hz; H_{ar}), 7.16 (d, 1H, *J* = 15.5 Hz; COCH=CH), 4.15 (t, 4H, *J* = 7.4 Hz; NH_2), 2.60 (s, 3H; $NCCH_3$), 2.57 (t, 2H, *J* = 7.4 Hz; CH_2CO), 2.37 (s, 3H; CH_3). ^{13}C NMR (101 MHz, $(CD_3)_2SO$, 25°C): δ = 180.20, 172.32, 170.84, 164.42, 161.95, 158.83, 140.67, 140.41, 138.24, 131.14, 130.87, 130.81, 127.07, 124.55, 122.10, 115.97, 115.76, 48.22, 32.28, 20.74,19.03.IR (KBr): ν = 1719, 1629, 1517 cm^{-1}. HRMS (ESI) for $C_{23}H_{21}FN_2O_3S$, $[M+H]^+$: 425.1257, found: 425.1337 $[M+H]^+$.

3-((5-(3-(4-Chlorophenyl)acryloyl)-4-methylthiazol-2-yl)(*p*-tolyl)amino)propanoic acid (26)

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.47 (br. s, 1H; OH), 7.75 (d, 2H, *J* = 8.4 Hz; H_{ar}), 7.51 (d, 1H, *J* = 15.5 Hz; COCH=CH), 7.44 (d, 2H, *J* = 8.1 Hz; H_{ar}), 7.35 (s, 4H; H_{ar}), 7,21 (d, 1H, *J* = 15.4 Hz; COCH=CH), 4.15 (t, 4H, *J* = 7.4 Hz; NH_2), 2.60 (s, 3H; $NCCH_3$), 2.57 (t, 2H, *J* = 7.4 Hz; CH_2CO), 2.37 (s, 3H; CH_3). ^{13}C NMR (101 MHz, $(CD_3)_2SO$, 25°C): δ = 180.07, 172.38, 170.91, 159.02, 140.64, 140.14, 138.23, 134.72, 133.45, 130.86, 130.22, 128,88, 127.06, 125.38, 122.06, 48.27, 32.34, 20.73,19.05. IR (KBr): ν = 1728, 1630, 1519 cm^{-1}. HRMS (ESI) for $C_{23}H_{21}ClN_2O_3S$, $[M+H]^+$: 441.0961; found: 441.1035 $[M+H]^+$.

3-((4-Methyl-5-(1-(2-phenylhydrazineylidene) ethyl)thiazol-2-yl)(*p*-tolyl)amino)propanoic acid (27)

A mixture of compound **23** (0.5 g, 1.4 mmol), phenylhydrazine (0.46 g, 4.2 mmol) and 6 drops of glacial acetic acid in methanol (30 mL) was refluxed for 5 h. Then the reaction mixture was cooled down to room temperature and diluted with water (30 mL). The precipitate was filtered off, washed with water, dried and purified by dissolving it in 5% aqueous sodium carbonate solution (2.5 g Na_2CO_3, 47.5 mL H_2O), filtering the solution and acidifying the filtrate with glacial acetic acid to pH 6. The obtained precipitate was filtered off, washed with water and dried to provide a light-yellow solid (0.54 g, 90%), m.p.: 210–211°C.

^{1}H NMR (400 MHz, $(CD_3)_2SO$, 25°C): δ = 12.27 (s, 1H; OH), 9.02 (s, 1H; NH), 7.30 (s, 4H; H_{ar}), 7.14 (t, *J* = 7.7 Hz, 2H; H_{ar}), 6.99 (d, *J* = 8.0 Hz, 2H; H_{ar}), 6.67 (t, *J* = 7.3 Hz, 1H; H_{ar}), 4.06 (t, *J* = 7.3 Hz, 2H; NCH_2), 2.58 (t, *J* = 7.3 Hz, 2H; CH_2CO), 2.39 (s, 3H; CH_3), 2.35 (s, 3H; CH_3), 2.17 (s, 3H; CH_3) ppm. ^{13}C NMR (101 MHz, $(CD_3)_2SO$, 25°C): δ = 172.54, 165.80, 145.91, 144.74, 141.71, 137.81, 137.06, 130.55, 128.81, 126.93, 121.67, 118.44, 112.40, 47.84, 32.33, 20.69, 18.16, 15.69 ppm. IR (KBr): ν = 1512, 1712, 3422 cm^{-1}. Elemental analysis calcd (%) for $C_{22}H_{24}N_4O_2S$: C 64.68, H 5.92, N 13.71; found: C 64.65, H 5.90, N 13.66.

Bacteria strains and culturing conditions.

The inhibitory activity evaluation of compounds

The antibacterial properties of the obtained compounds were evaluated against gram-positive strains of *Bacillus coagulans*, *Bacillus subtilis* and *Bacillus megaterium* and the gram-negative one *Escherichia coli* by the twofold serial dilution method [21]. A broad-spectrum antibiotic *Ampicillin* was used as a positive control. The *in vitro* antibacterial activity (MIC) of the antibiotic was 62.5 μg/mL for all strains.

RESULTS AND DISCUSSION

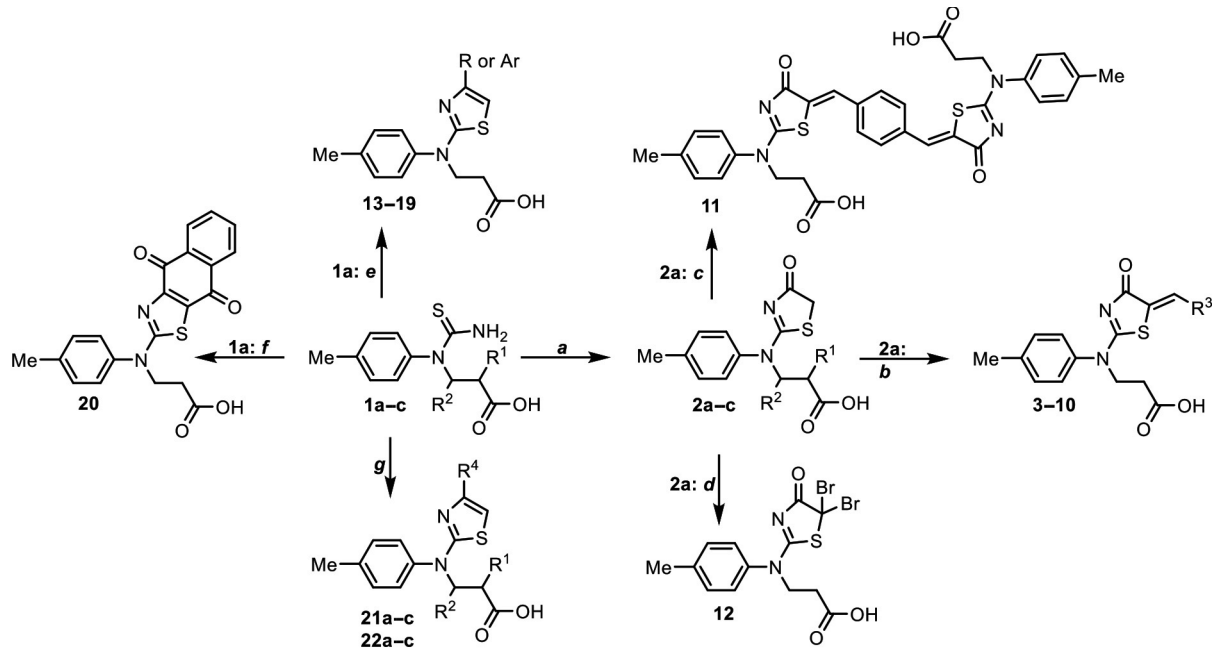
Synthesis

The synthesis was started from the preparation of thioureido acids **1a–c** according to the literature [22]. The obtained compounds were then applied for the formation of various Hantsch-thiazoles. Thiazolone **2a** was resynthesized [18], and **2b, c** were synthesized (Scheme 1) from the corresponding thioureido acid **1** and monochloroacetic acid. The reactions were carried out in water at reflux for 3 h and using a slight excess (1.2-fold) of sodium carbonate to avoid the formation of hydrogen halide. The structures of the formed compounds **2a–c** were established based on their NMR and IR spectroscopic and elemental analyses data. The characteristic peaks (^{1}H, at approx. 3.90 (^{1}H) and 40.0 (^{13}C) ppm for SCH_2, and the resonance at approx. 187.0 ppm for N–C=O) in their NMR spectra approve the formed thiazolone cycle. In the next step, several chemical transformations were made with **2a** by using the reactivity of the methylene fragment of the thiazolone ring. The condensation of compound **2a** with the appropriate aromatic aldehydes gave *Z*-configurated arylidene derivatives **3–10**. All reactions were performed in water at reflux in the presence of sodium carbonate and bifunctional catalyst glycine in the reaction mixture [23]. The ^{1}H NMR spectra of the obtained compounds revealed the disappearance of the singlet of SCH_2 proton and the presence of the addition spectral line of C=CH proton in the range of 7.51–7.85 ppm. The spectral lines of the carbon of the =CH group resonated in the aromatic area as additional resonances along with the resonances of newly attached phenyl fragment. The resonances of the carbons of C=N and C=O groups appeared to be characteristically shifted up-field and resonated at approx. 176.0 ppm (C=N) and 179.50 ppm (C=O) in comparison with 183.25 and 187.01 ppm for the same carbons of compound **2a**.

The terephthalaldehyde used instead of mono aldehydes was suitable to form bis thiazole derivative. Refluxing of compound **2a** with dialdehyde (2:1 equivalents) at the same reaction conditions as those of **3–10** led bis derivative **11**, the structure of which was confirmed by a detailed analysis of the ^{1}H NMR spectrum.

The bromination reaction of the thiazolone cycle of **2a** led to the generation of 5,5-dibromo analogue **12**. The reaction was accomplished using excess bromine (3.5 eq.) and stirring in acetic acid at 60°C for 4 h. The recorded spectra were in an excellent agreement with the target structure.

To obtain a series of thiazoles, thioureido acid **1a** was reacted with aromatic α-haloketones



Scheme 1. Synthesis of thiazole derivatives **2–22**.

1 and **2**: **a** $R^1 = R^2 = H$; **b** R^1 = Me, $R^2 = H$; **c** $R^1 = H$, R^2 = Me; **3** R^3 = Ph; **4** R^3 = 4-F-Ph; **5** R^3 = 4-Cl-Ph; **6** R^3 = 4-Br-Ph; **7** R^3 = 4-CH_3-Ph; **8** R^3 = 4-Me_2N-Ph; **9** R^3 = 4-O_2N-Ph; **10** R^3 = thien-2-yl; **13** R = H; **14** Ar = 4-F-Ph; **15** Ar = 3,4-diCl-Ph; **16** Ar = 4-O_2N-Ph; **17** Ar = 4-NC-Ph; **18** Ar = Naphthalen-2-yl; **19** Ar = 2-Chromenon-3-yl; **21a–c** R^4 = Me: **a** $R^1 = R^2 = H$; **b** R^1 = Me, $R^2 = H$; **c** $R^1 = H$, R^2 = Me; **22a–c** R^4 = Ph: **a** $R^1 = R^2 = H$; **b** R^1 = Me, $R^2 = H$; **c** $R^1 = H$, R^2 = Me.

Reagents and conditions: ***a*** monochloroacetic acid, Na_2CO_3 or AcONa, water, Δ, 2–3 h, AcOH to pH 6–7; ***b*** and ***c*** corresponding aldehyde (for **3–10**) or terephthalaldehyde (for **11**), Na_2CO_3, water, Δ, 3 h, 10% AcOH to pH 6; ***d*** AcOH, Br_2+AcOH, stirring, 60°C, 4 h, aq. AcONa, $Na_2S_2O_3$ to pH 7; ***e*** aq. 50% chloroacetaldehyde (**13**) or the corresponding ketone (**18**, **19**), dry acetone, Δ, 3 h (**13**) or 4 h (**18**, **19**), aq. AcONa, stirring, r.t., 1 h; ***e*** (for **14**, **16** and **17**) as described in Ref. [19]; ***e*** (for **15**) 2-bromo-3,4-dichloroacetophenone, AcONa, 2-PrOH, Δ, 3 h, water; ***f*** 2,3-dichloronaphthalene-1,4-dione, AcONa, dry acetone, Δ, 4 h, aq. 5% KOH, AcOH to pH 6–7; ***g*** chloroacetone or 2-bromoacetophenone, dry acetone, Δ, 4 h, water, AcONa, stirring, r.t. 10 min.

(compounds **14–19**), while the reaction with 50% aqueous chloroacetaldehyde solution gave 2-substituted thiazole **13**. The prepared thiazoles **13–19** were identified by their NMR, IR spectral and microanalysis data. The ^{1}H and ^{13}C NMR spectra of **13** showed two doublets at 6.68 and 7.17 ppm with J = 3.6 Hz (^{1}H) as well as 107.94 and 139.13 ppm (^{13}C) for SCH and NCH, respectively. Compounds **14**, **16** and **17** were resynthesized [19] and their spectra were in a good accordance with those of the reference. The structure of **15** was confirmed by the results of spectral and elemental analyses.

The NMR spectra of the prepared compounds **18** and **19** exhibited an intense splitting of peaks in the aromatic field of the spectra which was caused by the attachment of the naphthalene and 2-oxochromene moieties. The resonances of protons of the SCH groups of **18** and **19** in the ^{1}H NMR spectra are shifted down-field and appears at 8.41 and 8.64 ppm, respectively. A distinct one pot protocol toward the synthesis of fused tricyclic compound has been performed applying compound **1a** and 2,3-dichloronaphthalene-1,4-dione as initial compounds. Heating a mixture in dry acetone at reflux in the presence of excess sodium acetate afforded 3-((4,9-dioxo-4,9-dihydronaphtho[2,3-d] thiazol-2-yl)(*p*-tolyl)amino)propanoic acid **(20)**. The presence of a fused structure is evident from the NMR spectra of the compound. As for instance, the ^{13}C NMR spectrum of the isolated product **20** showed characteristic resonances at 172.13, 175.62, 175.89 and 180.01 ppm for three carbonyl groups of a new molecule.

To evaluate the impact of methyl groups at the α- and β-position of the β-alanine fragment on the biological efficacy of the thiazole derivatives **21a–c** and **22a–c** were synthesized from the initial compounds **1a–c** with the methyl (compounds **21a–c**) or phenyl (derivatives **22a–c**) fragment at the 4th position of the thiazole cycle. The appropriate thioureido acid **1**

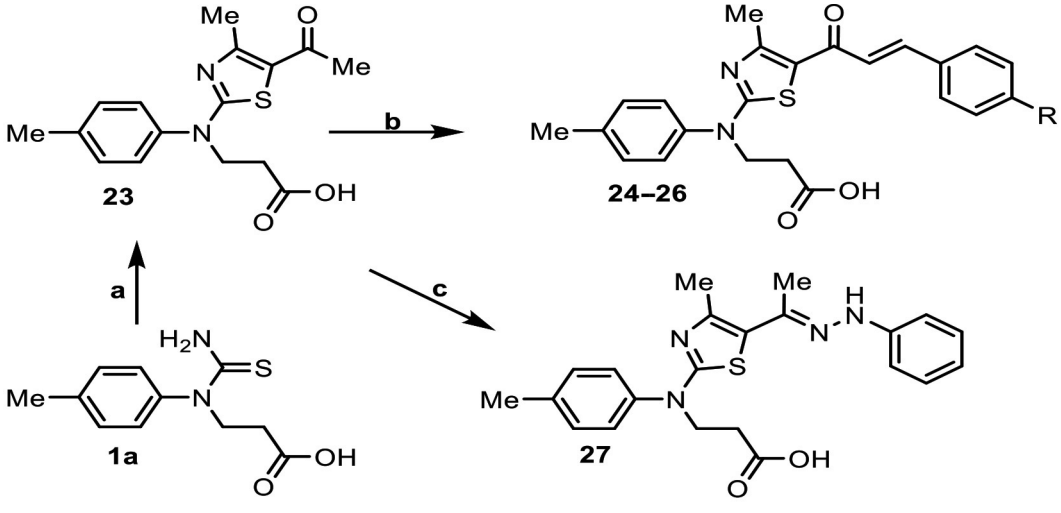
upon the reaction with chloroacetone or 2-bromoacetophenone in refluxing acetone provided hydrochloride salts which then were transferred into the base form with sodium acetate. The next step of the study was the Hantsch procedure to deliver 5-acetyl-4-methylthiazole **23** (Scheme 2) suitable for the further transformation to chalcone-type compounds. Condensation of acid **1a** with 3-chloropentane-2,4-dione and the subsequent Claisen–Schmidt cross-aldol condensation followed by the dehydration reaction resulted in compounds **24–26** containing a phenylacryloyl core in excellent yields (92–98%). The $C_\alpha = C_\beta$ double bond in the chalcone enon moiety determines the possibility of *Z* or *E* isomers. The ^{1}H NMR spectra revealed two doublets of the protons of the newly attached CO–CH=CH–Ph fragment (7.20 and 7.53 ppm for **24**, 7.16 and 7.53 ppm for **25** as well as 7.21 and 7.51 ppm for **26**), with the coupling constant $J > 15$ Hz indicating the *E* configuration of the compounds [24].

3-((4-methyl-5-(1-(2-phenylhydrazineylidene) ethyl)thiazol-2-yl)(*p*-tolyl)amino)propanoic acid (**27**) was prepared via condensation of 5-acetyl-4-methylthiazole **23** with phenylhydrazine in methanol in the presence of a catalytic amount of glacial acetic acid. The NMR, IR and microanalysis of the compounds confirmed the expected structure. The found values of the elemental analysis were in an excellent agreement with the calculated ones, and the ^{1}H NMR spectrum displayed characteristic singlets at 2.17 and 9.02 ppm for $CH_3C{=}N$ and NNH, respectively. The phenyl protons of the $C(CH_3){=}NNHPh$ fragment arise in the aromatic field as triplet (6.67 ppm, J = 7.3 Hz) integrated for one proton, and doublet (6.99 ppm, J = 8.0 Hz) and triplet (7.14 ppm, J = 7.7 Hz) each of which is integrated for two protons. The ^{13}C NMR spectrum revealed a resonance line at 15.69 ppm for CCH_3 and an increase in seven additional spectral lines attributable to the carbons of the C=NNHPh moiety in the aromatic field of the spectrum.

Biology

Minimum inhibition concentration (MIC) tests are widely applied for the evaluation of the lowest concentration of a compound at which it inhibits the growth of the bacteria. Generally, MIC antibacterial assays are performed by a two-fold microdilution procedure in which serial dilutions of a compound are made to produce different concentrations [25]. This allows the inhibition to be evaluated as a function of different concentrations of the test compound and to estimate antibacterial inhibition potential of the compound.

The synthesized thiazole derivatives were investigated for the antibacterial activity against three gram-positive *Bacillus* species such as *B. subtilis*, *B. coagulans* and *B. megaterium* and gram-negative bacteria *E. coli*. The data of the initial research



Scheme 2. Synthesis and transformation of 5-acetyl-4-methyltiazole derivative **23**.

24 R = H; **25** R = F; **26** R = Cl.

Reagents and conditions: ***a*** 3-chloropentane-2,4-dione, acetone Δ, 4 h, aq. AcONa, stirring, r.t. 15 min; ***b*** aq. 30% NaOH, 80°C, corresponding aromatic aldehyde, 2 h, saturated aq. NaCl; ***c*** phenylhydrazine, AcOH, MeOH, Δ, 5 h, water.

provided very promising information about the inhibitory properties of the synthesized compounds (Table).

The most sensitive strain appeared to be *Bacillus coagulans*, the growth of which was inhibited by the 15.63 μg/mL concentration of compounds **13** and **18** with an unsubstituted and 4-naphthyl-substituted thiazole ring, while the positive impact of ampicillin to that strain was determined to be 62.5 μg/mL. The incorporation of a 4-fluorophenyl substituent in the thiazole ring slightly reduced the MIC of the compound, but still remained 2-fold higher than that of the control. Other four 4-substituted thiazoles, namely compounds **15–17** and **19**, demonstrated activity equivalent to the control ampicillin. Obviously, the introduction of substituent containing electron-withdrawing groups (dichloro, nitro and ciano for **15**, **16** and **17**, respectively) at the 4th position of the thiazole ring decreases the inhibitory properties of the compounds, but still, they remain highly potent with the MIC identical to ampicillin. The same properties were shown by compound **19**, having a 2-chromenon-3-yl moiety in the molecule.

It is noteworthy that the introduction of 4-phenyl substitution in the thiazole bearing

Table. **Antibacterial activity of the synthesized compounds (MIC, μg/mL)**

Compound	Gram-positive bacteria			Gram-negative bacteria
	B. coagulans	*B. subtilis*	*B. megaterium*	*E. coli*
2a	125	125	62.5	250
2b	125	250	250	250
2c	250	250	250	250
3	250	250	250	125
4	125	125	125	250
5	250	250	250	125
8	250	125	250	250
9	250	62.5	125	125
10	125	250	250	250
11	125	125	250	250
12	250	250	250	250
13	15.63	125	125	250
14	31.25	250	125	250
15	62.5	250	125	250
16	62.5	250	125	250
17	62.5	250	62.5	250
18	15.63	250	125	250
19	62.5	250	125	125
20	125	250	125	250
21a	250	250	250	250
21b	250	250	250	125
22a	250	250	250	125
22b	250	125	250	125
22c	62.5	125	250	250
23	250	250	125	250
24	31.25	250	250	250
25	250	250	250	250
26	125	250	250	250
Ampicillin	62.5			

β-methylsubstituted β-alanine scaffold highly increased the antibacterial properties (MIC, 62.5 μg/mL) against *B. coagulans* in comparison with other compounds from the same set.

The biological properties of chalcones, including antibacterial potency, have been widely discussed in the literature [26]. In this study, the antibacterial test revealed chalcone with a 5-cinnamoyl fragment to be 2-fold more active (MIC, 31.25 μg/mL) than the conventional antibiotic, like ampicillin.

It should be noted that several compounds (**2a**, **9** and **17**) selectively inhibited some other gram-positive strains, namely *B. megaterium* (**2a**, **17**) and *B. subtilis* (**9**) with effects equivalent to the control antibiotic. The most resistant strain was found to be the gram-negative *Escherichia coli* the growth of which was inhibited by the compounds at MIC of 125 or 250 μg/mL.

CONCLUSIONS

In summary, we have synthesized a new library of thiazole derivatives by incorporating various substitutions in the 4th and 5th positions of the thiazole heterocycle as well and by including the unchanged amino acid fragment and its α-methyl and β-methyl analogues. The Hantsch method was applied for the preparation of thiazoles. The 5th position substituents were included by the condensation of the thiazolone with the appropriate aldehyde. The fused system derivative was obtained from thioureido acid by its reaction with 2,3-dichloronaphthalene-1,4-dione. Furthermore, the condensation of thioureido acid with 3-chloropentane-2,4-dione and the subsequent Claisen–Schmidt cross-aldol condensation followed by the dehydration reaction gave chalcone-type compounds. The attractive features of the applied reaction pathways are a simple procedure and isolation as well as the purification of target compounds, satisfactory to good yields of the obtained products, which makes it a suitable protocol for the synthesis of this class of compounds.

The antibacterial test results revealed that unsubstituted thiazole **13** and 4-naphthylthiazole **18** showed a profound and selective antibacterial activity targeting *Bacillus coagulans*. The MIC of these compounds was 4-fold higher than the clinically approved ampicillin. Compound **14** possessed 2-fold greater potency against this strain (MIC, 31.25 μg/mL) compared to ampicillin. The incorporation of electron withdrawing groups in the structure of thiazoles reduced their bioactivity but remained strong and comparable to the control antibiotic (MIC, 62.5 μg/mL). The introduction of 2-chromenon-3-yl fragment into the structure of the compound had the same effect on the inhibition properties. From the set of α- or β-methyl β-amino acid chain possessing thiazoles, 3-((4-phenylthiazol-2-yl)(*p*-tolyl)amino)butanoic acid (**22c**) demonstrated highly increased growth inhibition properties (MIC, 62.5 μg/mL) against *B. coagulans*. The modification of unsubstituted thiazole to a chalcone structure, namely to 5-cinnamoyl moiety bearing derivative, provided 2-fold higher inhibition (MIC, 31.25 μg/mL) than the conventional antibiotic ampicillin. Moreover, compounds **2a**, **9** and **17** were found to inhibit other gram-positive strains, such as *B. megaterium* (**2a**, **17**) and *B. subtilis* (**9**) with effects equivalent to the control antibiotic. Gram-negative *Escherichia coli*, which showed the highest resistance, was inhibited by the tested compounds only at MICs of 125 or greater at 250 μg/mL.

According to the test results, a series of aminothiazole derivatives based on **13** could be further studied as novel antibacterial agents targeting gram-positive bacteria strains.

Received 13 March 2023
Accepted 15 March 2023

References

1. G. S. S. Njateng, Zh. Du, D. Gatsing, et al., *BMC Complement. Altern. Med.*, **17**, 99 (2017).
2. S. Kumar, R. Aggawal, *Mini Rev. Org. Chem.*, **16**, 26 (2019).
3. S. H. Ali, A. R. Sayed, *Synth. Commun.*, **51**, 670 (2021).
4. K. M. Dawood, M. A. Raslan, A. A. Abbas, et al., *Front. Chem.*, **9**, 694870 (2021).
5. V. E. Borisenko, A. Koll, E. E. Kolmakov, A. G. Rjasnyi, *J. Mol. Struct.*, **2006**, 101 (2006).
6. K. A. Scott, J. T. Njardarson, *Top Curr. Chem.*, **376**, 5 (2018).
7. S. Pola, in: R. Varala (ed.), *Scope of Selective Heterocycles from Organic and Pharmaceutical Perspective*, p. 1–47, Rijeka, Croatia (2016).
8. N. Patel, Kh. Pandya, A. Gajjar, *Curr. Pharm. Anal.*, **12**, 275 (2016).
9. T. M. Potewar, S. A. Ingale, K. V. Srinivasan, *Tetrahedron*, **63**, 11066 (2007).

10. S. Rubino, R. Busà, A. Attanzio, et al., *Bioorg. Med. Chem.*, **25**, 2378 (2017).
11. S. M. Gomha, M. R. Abdelaziz, N. A. Kheder, H. M. Abdelaziz, S. Alterary, Y. N. Mabkhot, *Chem. Cent. J.*, **11**, 105 (2017).
12. M. Stasevych, V. Lubenets, R. Musyanovych, et al., *Chem. Het. Comp.*, **47**, 1050 (2011).
13. B. Grybaitė, R. Vaickelionienė, M. Stasevych, et al., *ChemistrySelect*, **4**, 6965 (2019).
14. B. Grybaitė, R. Vaickelionienė, M. Stasevych, O. Komarovska-Porokhnyavets, V. Novikov, V. Mickevičius, *Heterocycles*, **96**, 86 (2018).
15. I. Parašotas, K. Anusevičius, I. Jonuškienė, V. Mickevičius, *Chemija*, **25**, 107 (2014).
16. V. Mickevičius, A. Voskienė, I. Jonuškienė, et al., *Molecules*, **18**, 15000 (2013).
17. I. Vaškevičienė, V. Paketurytė, A. Zubrienė, K. Kantminienė, V. Mickevičius, D. Matulis, *Bioorg. Chem.*, **75**, 16 (2017).
18. M. Stasevych, V. Lubenets, R. Musyanovych, et al., *Chem. Heterocycl. Compd.*, **47** 1050 (2011).
19. B. Grybaitė, R. Vaickelionienė, M. Stasevych, O. Komarovska-Porokhnyavets, V. Novikov, V. Mickevičius, *Heterocycles*, **96**, 86 (2018).
20. B. Grybaitė, R. Vaickelionienė, M. Stasevych, et al., *ChemistrySelect* **4**, 6965 (2019).
21. M. Balouiri, M. Sadiki, S. K. Ibnsouda, *J. Pharm. Anal.*, **6**, 71 (2016).
22. R. Baltrušis, Z.-J. Beresnevičius, J. Vizgaitis, *Chem. Heterocycl. Compd.*, **17**, 816 (1981).
23. M. M. Chowdhry, D. M. P. Mingos, A. J. P. White, D. J. Williams, *J. Chem. Soc., Perkin Trans.*, **1**, 3495 (2000).
24. K. Liaras, A. Geronikaki, J. Glamoclija, A. Ciric, M. Sokovic, *Bioorg. Med. Chem.*, **19**, 3135 (2011).
25. J. F. S. Dos Santos, S. R. Tintino, Th. S. de Freitas, et al., *Comp. Immunol. Microbiol. Infect. Dis.*, **57**, 22 (2018).
26. J. S. Dhaliwal, S. Moshawih, Kh. W. Goh, et al., *Molecules*, **27**, 7062 (2022).

Birutė Sapijanskaitė-Banevič, Birutė Grybaitė, Rita Vaickelionienė, Ingrida Bružaitė

3-(TIAZOL-2-IL(*p*-TOLIL)AMINO)PROPANO RŪGŠTIES DARINIŲ SINTEZĖ, KITIMAI IR PIRMINIAI BIOLOGINIAI TYRIMAI

S a n t r a u k a

Šiame darbe atlikta įvairių 3-((4,5-dipakeistų tiazol-2-il)(*p*-tolil)amino)propano rūgšties darinių sintezė. Buvo susintetinta nauja tiazolo darinių biblioteka, įtraukiant įvairius pakaitus į 4- ir 5-tiazolo heterociklo padėtis ir išlaikant nepakeistą β-alanino fragmentą arba inkorporuojant jo α-metilo ir β-metilo analogus. Tiazolams gauti taikytas *Hantsch* tiazolų sintezės metodas. 5-osios tiazolo žiedo padėties pakaitai buvo įtraukti kondensuojant tiazoloną su atitinkamu aldehidu. Tiazolnaftochinono kondensuotoji sistema gauta tioureido rūgščiai reaguojant su 2,3-dichlornaftalen-1,4-dionu. Plečiant darinių įvairovę buvo atlikta tioureido rūgšties kondensacija su 3-chlorpentan-2,4-dionu ir vėlesnė *Claisen-Schmidt* kryžminė aldolio kondensacija, po kurios įvykus dehidratacijos reakcijai gauti chalkono tipo junginiai. Visi susintetinti junginiai identifikuoti remiantis ^{1}H, ^{13}C BMR, IR, masių spektroskopijos ir elementinės analizės duomenimis. Pasirinktų reakcijos metodų patrauklumas – paprastos metodikos, tikslinių junginių išskyrimas bei išgryninimas, turintis įtakos geroms gautų produktų išeigoms, įrodo jų tinkamumą šios klasės junginių sintezei.

Buvo atlikti pirminiai antibakteriniai susintetintų tiazolo darinių tyrimai prieš gram-teigiamas *Bacillus coagulans*, *Bacillus subtilis* ir *Bacillus megaterium* bakterijas bei gram-neigiamą *Escherichia coli* bakterijų padermę. Tyrimai atlikti dvigubo mikroskiedimo metodu, įvertinant minimalią slopinamąją koncentraciją (μg/ml). Antibakterinis testavimas atskleidė, kad darinys **13**, turintis nepakeistą tiazolo žiedą, ir 4-naftiltiazolas **18** selektyviai stipriai slopino *Bacillus coagulans* bakterijas. Jų minimali slopinamoji koncentracija (MIK) buvo keturis kartus stipresnė (MIK, 15,63 μg/ml) negu kliniškai patvirtinto ampicilino. 4-fluorfenilpakaitą turinčio tiazolo **14** MIK buvo šiek tiek silpnesnė (MIK, 31,25 μg/ml), tačiau visgi dar du kartus viršijo ampicilino poveikį šiai padermei. Į tiazolo žiedą inkorporavus elektrono akceptorines grupes turinčius fragmentus, gautų junginių bioaktyvumas susilpnėjo, tačiau jų minimali slopinamoji koncentracija prilygo kontroliniam antibiotikui (MIK, 62,5 μg/ml). Reikia paminėti, kad 2-chromenon-3-ilfragmento įvedimas į junginio struktūrą (junginys **19**) inicijavo tokį pat slopinamąjį poveikį, kaip ir elektrono akceptorinių grupių buvimas. Tiriant α- ir β-metil-β-aminorūgščių fragmentus turinčių tiazolų antibakterines savybes nustatyta, kad 3-((4-feniltiazol-2-il)(*p*-tolil)amino)butano rūgštis (**22c**) *B. coagulans* padermės augimą slopino taip pat (MIK, 62,5 μg/ml) kaip ir ampicilinas. Nepakeistą tiazolo žiedą modifikavus į chalkono tipo darinį, turintį 5-cinamoilo fragmentą, junginys įgavo stiprų slopinamąjį poveikį, net dvigubai (MIK, 31,25 μg/ml) stipresnį nei įprastai naudojamas antibiotikas ampicilinas. Taip pat pastebėta, kad junginiai **2a**, **9** ir **17** puikiai slopina ir kitas gram-teigiamas bakterijų padermes: **2a** ir **17** – *B. megaterium*, o **9** – *B. subtilis*. Jų poveikis prilygo kontrolinio antibiotiko efektui. Tyrimo metu nustatyta, kad didžiausią atsparumą tirtiems tiazolams parodė gram-neigiamos *Escherichia coli* bakterijos. Jų augimui slopinti prireikė 125 arba net 250 μg/mL koncentracijų junginių tirpalų.

Atsižvelgiant į biologinių tyrimų rezultatus, aminotiazolo darinių, suformuotų 3-(1,3-tiazol-2-il(*p*-tolil)amino)propano rūgšties **(13)** pagrindu, serija gali būti toliau tiriama kuriant tikslinius, prieš gram-teigiamas bakterijų padermes nukreiptus antibakterinius preparatus.