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Synthesis and antibacterial activity of new *N*-substituted 7-amino-4-methyl-2*H*-chromen-2-ones

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Abstract *N*-Substituted 7-amino-4-methyl-2*H*-chromen-2-ones containing one or two functionalized azole or azine moieties were synthesized. The structures of all synthesized compounds were confirmed by IR, ^{1}H NMR, and ^{13}C NMR spectroscopy. Some of the synthesized compounds exhibited weak antibacterial activity against *Rhizobium radiobacter*, *Escherichia coli*, and *Xanthomonas campestris*.

Keywords Pyrrolidin-2-ones · Azoles · Antibacterial activities · Chromen-2-one

Introduction

Biologically active compounds possessing antibacterial [1–5], antiviral [5–7], anticancer [8, 9], antioxidant [10, 11], etc. activities have been found among 2*H*-chromen-2-one derivatives. Furthermore, 2*H*-chromen-2-ones have been characterized by fluorescence properties, and therefore, are applied in the production of products able to glow on fabric or paper (markers, laser dyes, and optical brighteners) and in the manufacture of molecular diodes [12–14]. Pyrrolidin-2-one compounds are considered to be important chemical precursors of various physiologically active compounds and pharmaceutical agents. They possess a variety of biological activities such as antibacterial [15–17], anticancer [18–20], anti-HIV-1, anticonvulsant [21, 22], and have been indicated as ketoamide-based cathepsin K inhibitors [23] and agonists of human melanocortin-4 receptor [24].

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Thiazole is another important pharmacophore associated with various biological activities such as antimicrobial [25–29], antifungal [30, 31], antiviral [32, 33], anticancer [34, 35], anti-inflammatory [36–38], antidepressant [39], and antidiabetic [40].

In view of these observations, we report on the synthesis of new 2*H*-chromen-2-one derivatives with functionalized thiazole and pyrrolidinone moieties and investigation of their antibacterial activity.

Experimental section

Chemistry

General methods

The melting points were determined on a MEL-TEMP (Electrothermal, Bibby Scientific Company, Burlington, NJ, USA) melting point apparatus and are uncorrected. IR spectra (ν, cm^{-1}) were recorded on a Perkin–Elmer Spectrum BX FT–IR spectrometer using KBr tablets. The ^{1}H and ^{13}C-NMR spectra were recorded in DMSO-d_6 on a Varian Unity Inova (300, 75 MHz), Brucker Avance III (400, 101 MHz), and Brucker Avance III (700, 176 MHz) spectrometers. Chemical shifts (δ) are reported in parts per million (ppm) calibrated from TMS (0 ppm) as an internal standard for ^{1}H NMR, and DMSO-d_6 (39.43 ppm) for ^{13}C NMR. Elemental analyses (C, H, N) were performed on an Elemental Analyzer CE-440 (Exeter Analytical, Inc., North Chelmsford, MA, USA). The reaction course and the purity of the synthesized compounds were monitored by TLC using aluminium plates pre-coated with silica gel 60 F_{254} (MerckKGaA, Darmstadt, Germany). Reagents were purchased from Sigma-Aldrich (St. Louis, MO, USA) and Fluka (Buchs, Switzerland).

7-Amino-4-methyl-2H-chromen-2-one (1) was prepared as described in [41]

*1-(4-Methyl-2-oxo-2H-chromen-7-yl)-5-oxopyrrolidine-3-carboxylic acid (**2**)*

The mixture of 7-amino-4-methyl-2*H*-chromen-2-one (**1**) (4.28 g, 24 mmol) and itaconic acid (5.12 g, 40 mmol) was heated at 155–160 °C for 5 h. Then the reaction mixture was cooled to room temperature and dissolved in 10 % aqueous NaOH solution (50 mL). The solution was filtered off, and the filtrate was acidified with hydrochloric acid to pH 2. The formed precipitate was filtered off, washed with water, dried, and recrystallized from methanol to afford light-brown solid, yield 6.16 g (89 %), mp 234–235 °C; IR (KBr): 3084 (OH), 1729, 1720, 1671 (3C=O) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 2.40 (*d*, $J = 1.2$ Hz, 3H, CH_3), 2.49–2.53 (*m*, 2H, CH_2CO), 3.28–3.46 (*m*, 1H, C*H*CH_2), 3.99–4.17 (*m*, 2H, NCH_2), 6.29 (*d*, $J = 1.2$ Hz, 1H, CC*H*CO), 7.65–7.80 (*m*, 3H, H_{ar}), 12.83 (br. *s*, 1H, OH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ 17.8 (CH_3), 34.8 (*C*HCH_2), 35.3 (*C*H_2CO), 49.8 (NCH_2), 105.8, 112.7, 114.8, 115.3, 125.6, 142.0, 152.8, 153.3 (C_{ar},

C*C*HCO), 159.8, 172.6, 173.9 (3C=O) ppm. Anal. Calcd. for $C_{15}H_{13}NO_5$: C, 62.72; H, 4.56; N, 4.88 %. Found: C, 62.90; H, 4.76; N, 4.73 %.

*Methyl 1-(4-methyl-2-oxo-2H-chromen-7-yl)-5-oxopyrrolidine-3-carboxylate (**3**)*

A mixture of compound **2** (3.44 g, 12 mmol), methanol (50 mL), and sulfuric acid (5 mL) was heated at reflux for 6 h. The liquid fractions were evaporated under reduced pressure and the residue was poured over with 10 % aqueous Na_2CO_3 solution (150 mL). The formed precipitate was filtered off, washed with water, dried, and recrystallized from propan-2-ol to afford white solid, yield 3.26 g (90 %), mp 157–158 °C; IR (KBr): 1736, 1717, 1616 (3C=O), 1034 (O–C) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 2.40 (*d*, $J = 1.2$ Hz, 3H, CH_3), 2.73–2.93 (*m*, 2H, CH_2CO), 3.43–3.56 (*m*, 1H, C*H*CH_2), 3.69 (*s*, 3H, OCH_3), 3.99–4.18 (*m*, 2H, NCH_2), 6.28 (*d*, $J = 1.2$ Hz, 1H, CC*H*CO), 7.63–7.76 (*m*, 3H, H_{ar}) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ 17.8 (CH_3), 34.6 (*C*HCH_2), 35.1 (*C*H_2CO), 49.6 (NCH_2), 52.1 (OCH_3), 105.8, 112.7, 114.8, 115.4, 125.6, 141.8, 152.8, 153.2 (C_{ar}, *CC*HCO), 159.8, 172.3, 172.8 (3C=O) ppm. Anal. Calcd. for $C_{16}H_{15}NO_5$: C, 63.78; H, 5.02; N, 4.65 %. Found: C, 63.96; H, 5.09; N, 4.53 %.

*1-(4-Methyl-2-oxo-2H-chromen-7-yl)-5-oxopyrrolidine-3-carbohydrazide (**4**)*

A mixture of compound **3** (3.01 g, 10 mmol), hydrazine monohydrate (1.50 g, 30 mmol), and propan-2-ol (15 mL) was heated at reflux for 1 h. Then the reaction mixture was cooled to room temperature, the formed precipitate was filtered off and washed with propan-2-ol to give white solid, yield 2.65 g (88 %), mp 225–226 °C; IR (KBr): 3267, 3107 (NH, NH_2), 1710, 1689, 1616 (3C=O) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 2.40 (*d*, $J = 1.0$ Hz, 3H, CH_3), 2.60–2.88 (*m*, 2H, CH_2CO), 3.13–3.26 (*m*, 1H, C*H*CH_2), 3.85–4.10 (*m*, 2H, NCH_2), 4.35 (br. *s*, 2H, NH_2), 6.28 (*d*, $J = 1.2$ Hz, 1H, CH_2CO), 7.64–7.79 (*m*, 3H, H_{ar}), 9.31 (*s*, 1H, NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ 17.8 (CH_3), 33.8 (C*H*CO), 35.9 (*C*H_2CO), 50.5 (NCH_2), 105.6, 112.6, 114.7, 115.3, 125.6, 142.0, 152.8, 153.3 (C_{ar}, *CC*HCO), 159.9, 171.2, 172.9 (3C=O) ppm. Anal. Calcd. for $C_{15}H_{15}N_3O_4$: C, 59.80; H, 5.02; N, 13.95 %. Found: C, 59.61; H, 5.23; N, 13.76 %.

General procedures for the synthesis of 1,3-thiazoles 5–9

A mixture of a corresponding substituted α-haloketone (2.5 mmol), potassium thiocyanate (0.29 g, 3 mmol), and ethanol (15 mL) was heated at 50–60 °C for 4 h. Then 7-amino-4-methyl-2*H*-chromen-2-one (**1**) (0.44 g, 2.5 mmol) was added to the reaction mixture and the heating at reflux was continued for 40 h. The reaction mixture was cooled to room temperature, the formed precipitate was filtered off, washed with propan-2-ol, and dried. Unreacted 7-amino-4-methyl-2*H*-chromen-2-one (**1**) was dissolved in 10 % hydrochloric acid (20 mL), whereas the formed target 1,3-thiazole **5–9** was filtered off, washed with water, dried, and recrystallized from methanol.

*4-Methyl-7-[(4-phenyl-1,3-thiazol-2-yl)amino]-2H-chromen-2-one (**5**)*

Light-brown solid, yield 0.52 (62 %), mp 259–260 °C; IR (KBr): 3283 (NH), 1691 (C=O), 1626 (C=N) cm^{-1}; ^{1}H NMR (400 MHz, DMSO-d_6): δ 2.38 (*d*, $J = 1.0$ Hz, 3H, CH_3), 6.18 (*d*, $J = 1.2$ Hz, 1H, CC*H*CO), 7.30–8.05 (*m*, 9H, H_{ar} and S–CH), 10.86 (*s*, 1H, NH) ppm; ^{13}C NMR (101 MHz, DMSO-d_6): δ 18.1 (CH_3), 102.8 (S–CH), 104.5, 111.0, 113.2, 113.4, 125.7, 126.1, 127.9, 128.8, 134.3, 144.3, 150.3, 153.3, 154.4 (C_{ar}, *C*CHCO, and CH*C*N), 160.4 (C=O), 162.1 (S–C=N) ppm. Anal. Calcd. for $C_{19}H_{14}N_2O_2S$: C, 68.25; H, 4.22; N, 8.38 %. Found: C, 68.50; H, 4.36; N, 8.31 %.

*7-{[4-(4-Chlorophenyl)-1,3-thiazol-2-yl]amino}-4-methyl-2H-chromen-2-one (**6**)*

Yellow solid, yield 0.63 (68 %), mp 292–293 °C; IR (KBr): 3294 (NH), 1705 (C=O), 1627 (C=N) cm^{-1}; ^{1}H NMR (700 MHz, DMSO-d_6): δ 2.40 (*s*, 3H, CH_3), 6.19 (*s*, 1H, CC*H*CO), 7.44–8.03 (*m*, 8H, H_{ar} and S–CH), 10.85 (*s*, 1H, NH) ppm; ^{13}C NMR (176 MHz, DMSO-d_6): δ 18.0 (CH_3), 102.8 (S–CH), 105.3, 111.3, 113.3, 126.1, 127.3, 128.8, 132.2, 133.1, 144.1, 144.0, 149.1, 153.3, 154.3 (C_{ar}, *C*CHCO, and CH*C*N), 160.3 (CO), 162.3 (SCN) ppm. Anal. Calcd. for $C_{19}H_{13}ClN_2O_2S$: C, 61.87; H, 3.55; N, 7.60 %. Found: C, 61.64; H, 3.63; N, 7.74 %.

*4-Methyl-7-{[4-(4-nitrophenyl)-1,3-thiazol-2-yl]amino}-2H-chromen-2-one (**7**)*

Dark yellow solid, yield 0.56 (59 %), mp > 320 °C; IR (KBr): 3300 (NH), 1694 (C=O), 1629 (C=N) cm^{-1}; ^{1}H NMR (400 MHz, DMSO-d_6): δ 2.40 (*s*, 3H, CH_3), 6.21 (*s*, 1H, CC*H*CO), 7.36–8.46 (*m*, 8H, H_{ar} , and S–CH), 10.95 (*s*, 1H, NH) ppm; ^{13}C NMR (101 MHz, DMSO-d_6): δ 18.1 (CH_3), 103.0 (S–CH), 109.4, 111.2, 113.6, 124.3, 126.5, 140.2, 142.2, 144.0, 146.4, 146.5, 153.3, 154.3 (C_{ar}, *C*CHCO, and CH*C*N), 160.3 (C=O), 162.7 (S–C=N) ppm. Anal. Calcd. for $C_{19}H_{13}N_3O_4S$: C, 60.15; H, 3.45; N, 11.08 %. Found: C, 60.28; H, 3.54; N, 11.35 %.

*7-{[4-(4-Bromophenyl)-1,3-thiazol-2-yl]amino}-4-methyl-2H-chromen-2-one (**8**)*

Dark yellow solid, yield 0.67 (65 %), mp 280–281 °C; IR (KBr): 3295 (NH), 1706 (C=O), 1626 (C=N) cm^{-1}; ^{1}H NMR (400 MHz, DMSO-d_6): δ 2.39 (*d*, $J = 0.8$ Hz, 3H, CH_3), 6.19 (*d*, $J = 1.1$ Hz, 1H, CC*H*CO), 7.46–7.98 (*m*, 8H, H_{ar} , and S–CH), 10.87 (*s*, 1H, NH) ppm; ^{13}C NMR (101 MHz, DMSO-d_6): δ 18.1 (CH_3), 102.9 (S–CH), 105.5, 111.1, 113.3, 113.5, 120.9, 126.2, 127.6, 131.8, 133.5, 144.1, 149.1, 153.3, 154.4 (C_{ar}, *C*CHCO, and CH*C*N), 160.3 (C=O), 162.3 (S–C=N) ppm. Anal. Calcd. for $C_{19}H_{13}BrN_2O_2S$: C, 55.22; H, 3.17; N, 6.78 %. Found: C, 55.44; H, 3.33; N, 6.89 %.

*4-Methyl-7-{[4-(2-oxo-2H-chromen-3-yl)-1,3-thiazol-2-yl]amino}-2H-chromen-2-one (**9**)*

Green solid, yield 0.54 g (54 %), mp > 320 °C; IR (KBr): 3272 (NH), 1708 (2C=O), 1619 (C=N) cm^{-1}; ^{1}H NMR (700 MHz, DMSO-d_6): δ 2.42 (*s*, 3H, CH_3),

6.21 (*d*, $J = 1.1$ Hz, 1H, CC*H*CO), 7.37–7.98 (*m*, 9H, H_{chrom} and S–CH), 8.66 (*s*, 1H, NCCCH), 10.87 (*s*, 1H, NH) ppm; ^{13}C NMR (176 MHz, DMSO-d_6): δ 18.0 (CH_3), 103.1 (S–CH), 111.2, 111.3, 113.5, 113.6, 115.9, 119.2, 120.2, 124.8, 126.3, 128.9, 131.8, 138.7, 143.8, 144.0, 152.4, 153.2, 154.2 (C_{chrom}, *C*CHCO, and CH*C*N), 158.8, 160.3 (2C=O), 161.7 (S–C=N) ppm. Anal. Calcd. for $C_{22}H_{14}N_2O_4S$: C, 65.66; H, 3.51; N, 6.96 %. Found: C, 65.47; H, 3.67; N, 6.83 %.

*1-(4-Methyl-2-oxo-2H-chromen-7-yl)-5-oxo-N-phenylpyrrolidine-3-carboxamide (**11**)*

A mixture of compound **2** (1.00 g, 3.5 mmol) and thionyl dichloride (2.5 mL) was heated at reflux for 3 h. The liquid fractions were evaporated under reduced pressure. Aniline (0.49 g, 5.25 mmol) and toluene (10 mL) were added to the residue, and the reaction mixture was heated at reflux for 2 h. Then it was cooled to room temperature, the formed precipitate was filtered off, washed with diethyl ether, dried, and recrystallized from methanol to afford white solid, yield 0.61 g (48 %), mp 189–190 °C; IR (KBr): 3063 (NH), 1717, 1684, 1653 (3C=O) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 2.41 (*d*, $J = 0.8$ Hz, 3H, CH_3), 2.76–2.98 (*m*, 2H, CH_2CO), 3.44–3.58 (*m*, 1H, *CH*CH_2), 4.01–4.24 (*m*, 2H, NCH_2), 6.29 (*d*, $J = 1.1$ Hz, 1H, CCHCO), 7.02–7.84 (*m*, 8H, H_{ar}), 10.27 (*s*, 1H, NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ 17.8 (CH_3), 35.9 (*C*H_2CO), 35.9 (*C*HCH$_2$), 50.5 (NCH_2), 105.7, 112.7, 114.7, 115.3, 119.2, 123.4, 125.6, 128.6, 138.8, 142.0, 152.8, 153.3 (C_{ar}, *C*CHCO), 159.8, 170.7, 172.8 (3C=O) ppm. Anal. Calcd. for $C_{21}H_{18}N_2O_4$: C, 69.60; H, 5.01; N, 7.73 %. Found: C, 69.41; H, 4.74; N, 7.65 %.

*1-({[1-(4-Methyl-2-oxo-2H-chromen-7-yl)-5-oxopyrrolidin-3-yl]carbonyl}amino)-5-oxopyrrolidine-3-carboxylic acid (**12**)*

A mixture of hydrazide **4** (0.74 g, 2.5 mmol), itaconic acid (0.39 g, 3 mmol), and glacial acetic acid (10 mL) was heated at reflux for 36 h. Then it was cooled down to room temperature and diluted with water (20 mL). The precipitate formed was filtered off, washed with propan-2-ol, and recrystallized from methanol to afford grey solid, yield 0.76 g (74 %), mp 239–240 °C; IR (KBr): 3285 (OH), 3108 (NH), 1719 str., 1691, 1669, 1615 (5C=O) cm^{-1}; ^{1}H NMR (700 MHz, DMSO-d_6): δ 2.41 (*d*, $J = 1.2$ Hz, 3H, CH_3), 2.47–2.62 (*m*, 2H, (**b**) CH_2CO), 2.66–2.91 (*m*, 2H, (**a**) CH_2CO), 3.27–3.32 (*m*, 1H, (**b**) C*H*CH_2), 3.33–3.40 (*m*, 1H, (**a**) C*H*CH_2), 3.58–3.72 (*m*, 2H, (**b**) NCH_2), 3.91–4.16 (*m*, 2H, (**a**) NCH_2), 6.30 (*d*, $J = 1.2$ Hz, 1H, CCHCO), 7.66–7.79 (*m*, 3H, H_{ar}), 10.48 (*s*, 1H, NH), 12.70 (br. *s*, 1H, OH) ppm; ^{13}C NMR (176 MHz, DMSO-d_6): δ 18.0 (CH_3), 31.2 ((**b**) *C*H_2CO), 33.5 ((**a**) *C*H_2CO), 34.1((**a**) *C*HCH$_2$), 35.7 ((**b**) *C*HCH$_2$), 49.6 ((**a**) NCH_2), 50.3 ((**b**) NCH_2), 105.9, 112.8, 114.9, 115.5, 125.8, 142.0, 153.0, 153.4 (C_{ar}, *C*CHCO), 160.0, 170.8, 171.3, 172.7, 173.9 (5C=O) ppm. Anal. Calcd. for $C_{20}H_{19}N_3O_7$: C, 58.11; H, 4.63; N, 10.17 %. Found: C, 58.01; H, 4.78; N, 10.05 %.

General procedure for the preparation of hydrazones 13–19

A mixture of hydrazide **4** (0.74 g, 2.5 mmol), corresponding aldehyde (3 mmol), and 1,4-dioxane (20 mL) was heated at reflux for 6 h. Then it was cooled to room temperature. The precipitate formed was filtered off, washed with propan-2-ol, and recrystallized from 1,4-dioxane.

*1-(4-Methyl-2-oxo-2H-chromen-7-yl)-5-oxo-N′-[phenylmethylidene]pyrrolidine-3-carbohydrazide (**13**)*

White solid, yield 0.76 g (78 %), mp 236–237 °C; IR (KBr): 3075 (NH), 1716, 1692, 1676 (3C=O), 1613 (N=CH) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 2.39 (*s*, 3H, CH_3), 2.74–3.01 (*m*, 2H, CH_2CO), 3.99–4.27 (*m*, 3H, $CHCH_2$, NCH_2), 6.28 (*s*, 1H, CCHCO), 7.36–7.83 (*m*, 8H, H_{ar}), 8.04, 8.22 (2*s*, 0.7:0.3(1H), N=CH), 11.63, 11.70 (2*s*, 0.7:0.3(1H), NH) ppm. Anal. Calcd. for $C_{22}H_{19}N_3O_4$: C, 67.86; H, 4.92; N, 10.79 %. Found: C, 67.72; H, 5.09; N, 10.71 %.

*1-(4-Methyl-2-oxo-2H-chromen-7-yl)-5-oxo-N′-[4-methylphenylmethylidene] pyrrolidine-3-carbohydrazide (**14**)*

White solid, yield 0.74 g (73 %), mp 251–252 °C; IR (KBr): 3065 (NH), 1721, 1700, 1672 (3C=O), 1616 (N=CH) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 2.32 (*s*, 3H, PhC*H*$_3$), 2.40 (*s*, 3H, CH_3), 2.73–2.97 (*m*, 2H, CH_2CO), 3.98–4.26 (*m*, 3H, C*H*CH_2, NCH_2), 6.28 (*s*, 1H, CCHCO), 7.55–7.79 (*m*, 7H, H_{ar}), 8.00, 8.18 (2*s*, 0.6:0.4(1H), N=C*H*), 11.55, 11.63 (2*s*, 0.6:0.4(1H), NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ 17.9 (CH_3), 21.7 (Ph*C*H$_3$), 33.4 (*C*H_2CO), 36.0 (*C*HCO), 50.7 (NCH_2), 106.5, 113.5, 115.6, 116.1, 126.5, 127.6, 130.1, 132.1, 140.4, 142.8, 147.9, 153.7, 154.1, 160.7 (C_{ar}, *CC*HCO, N=CH), 169.1, 173.6, 174.0 (3C=O) ppm. Anal. Calcd. for $C_{23}H_{21}N_3O_4$: C, 68.47; H, 5.25; N, 10.42 %. Found: C, 68.68; H, 5.19; N, 10.25 %.

*1-(4-Methyl-2-oxo-2H-chromen-7-yl)-5-oxo-N′-[(4-methoxyphenyl)methylidene] pyrrolidine-3-carbohydrazide (**15**)*

White solid, yield 0.75 g (72 %), mp 239–240 °C; IR (KBr): 3197 (NH), 1712, 1680, 1690 (3C=O), 1616 (N=CH) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 2.40 (*s*, 3H, CH_3), 2.78–2.97 (*m*, 2H, CH_2CO), 3.79 (*s*, 3H, OCH_3), 3.99–4.26 (*m*, 3H, C*H*CH_2 and NCH_2), 6.29 (*d*, $J = 1.2$ Hz, 1H, CCHCO), 6.95–7.77 (*m*, 7H, H_{ar}), 7.99, 8.17 (2*s*, 0.6:0.4(1H), N=CH), 11.45, 11.52 (2*s*, 0.6:0.4(1H), NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ 17.9 (CH_3), 32.6 (*C*H_2CO), 35.1 (*C*HCO), 50.0 (NCH_2), 55.2 (OCH_3), 105.7, 105.8, 112.7, 114.2, 114.7, 114.8, 115.3, 125.6, 126.0, 128.4, 128.6, 142.1, 143.6, 146.9, 152.8, 153.3, 159.8 (C_{ar}, *CC*HCO, N=CH), 168.1, 172.8, 173.0 (3C=O) ppm. Anal. Calcd. for $C_{23}H_{21}N_3O_5$: C, 65.86; H, 5.05; N, 10.02 %. Found: C, 65.69; H, 5.10; N, 10.19 %.

*N′-[(2-Hydroxyphenyl)methylidene]-1-(4-methyl-2-oxo-2H-chromen-7-yl)-5-oxopyrrolidine-3-carbohydrazide (**16**)*

Brown solid, yield 0.68 g (67 %), mp 280–281 °C; IR (KBr): 3200 (OH), 3046 (NH), 1720, 1696, 1674 (3C=O), 1617 (N=CH) cm^{-1}; ^1H NMR (400 MHz, DMSO-d_6): δ 2.41 (*d*, $J = 1.9$ Hz, 3H, CH_3), 2.76–2.96 (*m*, 2H, CH_2CO), 4.02–4.25 (*m*, 3H, C*H*CH_2, NCH_2), 6.30 (*d*, $J = 1.2$ Hz, 1H, CCHCO), 6.81–7.80 (*m*, 7H, H_{ar}), 8.35, 8.43 (2*s*, 0.4:0.6(1H), N=CH), 10.07, 11.03, 11.55, 11.92 (4*s*, 0.4:0.6:0.4:0.6(2H), OH and NH) ppm; ^{13}C NMR (101 MHz, DMSO-d_6): δ 18.0 (CH_3), 32.7 (*C*H_2CO), 35.2 (*C*HCO), 50.1 (NCH_2), 105.8, 105.9, 112.8, 114.9, 115.0, 115.5, 116.1, 118.7, 119.4, 120.3, 125.8, 126.2, 129.1, 131.2, 131.5, 141.1, 142.1, 147.3, 153.0, 153.5, 156.4, 157.3, 160.0 (C_{ar}, *CC*HCO, N=CH), 168.3, 172.9, 173.1 (3C=O) ppm. Anal. Calcd. for $C_{22}H_{19}N_3O_5$: C, 65.18; H, 4.72; N, 10.37 %. Found: C, 65.32; H, 4.83; N, 10.42 %.

*N′-[(4-Chlorophenyl)methylidene]-1-(4-methyl-2-oxo-2H-chromen-7-yl)-5-oxopyrrolidine-3-carbohydrazide (**17**)*

White solid, yield 0.77 g (73 %), mp 276–277 °C; IR (KBr): 3196 (NH), 1719, 1698, 1679 (3C=O), 1616 (N=CH) cm^{-1}; ^1H NMR (400 MHz, DMSO-d_6): δ 2.41 (*s*, 3H, CH_3), 2.74–2.96 (*m*, 2H, CH_2CO), 4.01–4.26 (*m*, 3H, C*H*CH_2, NCH_2), 6.30 (*d*, $J = 1.2$ Hz, 1H, CCHCO), 6.81–7.80 (*m*, 7H, H_{ar}), 8.03, 8.21 (2*s*, 0.6:0.4(1H), N=CH), 11.68, 11.77 (2*s*, 0.6:0.4(1H), NH) ppm; ^{13}C NMR (101 MHz, DMSO-d_6): δ 18.0 (CH_3), 32.7 (*C*H_2CO), 35.2 (*C*HCO), 50.1 (NCH_2), 105.8, 105.9, 112.8, 114.9, 115.0, 115.5, 116.1, 118.7, 119.4, 120.3, 125.8, 126.2, 129.1, 131.2, 131.5, 141.1, 142.1, 147.3, 153.0, 153.5, 156.4, 157.3, 160.0 (C_{ar}, *CC*HCO, N=CH), 168.3, 172.9, 173.1 (3C=O) ppm. Anal. Calcd. for $C_{22}H_{18}ClN_3O_4$: C, 62.34; H, 4.28; N, 9.91 %. Found: C, 62.52; H, 4.37; N, 9.80 %.

*1-(4-Methyl-2-oxo-2H-chromen-7-yl)-N′-[(4-nitrophenyl)methylidene]-5-oxopyrrolidine-3-carbohydrazide (**18**)*

Light brown solid, yield 0.81 g (75 %), mp 304–305 °C; IR (KBr): 3200 (NH), 1731, 1685, 1669 (3C=O), 1616 (N=CH) cm^{-1}; ^1H NMR (400 MHz, DMSO-d_6): δ 2.41 (*s*, 3H, CH_3), 2.76–2.98 (*m*, 2H, CH_2CO), 4.03–4.28 (*m*, 3H, C*H*CH_2, NCH_2), 6.30 (*s*, 1H, CCHCO), 7.69–8.44 (*m*, 8H, H_{ar} and N=CH), 11.92, 12.00 (2*s*, 0.6:0.4(1H), NH) ppm; ^{13}C NMR (101 MHz, DMSO-d_6): δ 18.0 (CH_3), 32.7 (*C*H_2CO), 35.2 (*C*HCO), 50.0 (NCH_2), 106.0, 112.9, 115.0, 115.5, 124.1, 125.8, 127.9, 140.4, 141.5, 142.2, 147.7, 153.0, 153.4 (C_{ar}, *CC*HCO, N=CH), 160.0, 173.0, 173.8 (3C=O) ppm. Anal. Calcd. for $C_{22}H_{18}N_4O_6$: C, 60.83; H, 4.18; N, 12.90 %. Found: C, 60.97; H, 4.25; N, 12.77 %.

*N′-[(4-Bromophenyl)methylidene]-1-(4-methyl-2-oxo-2H-chromen-7-yl)-5-oxopyrrolidine-3-carbohydrazide (**19**)*

White solid, yield 0.78 g (67 %), mp 295–296 °C; IR (KBr): 3109 (NH), 1727, 1681, 1679 (3C=O), 1619 (N=CH) cm^{-1}; ^1H NMR (400 MHz, DMSO-d_6): δ 2.41 (*d*, $J = 1.1$ Hz, 3H, CH_3), 2.75–2.99 (*m*, 2H, CH_2CO), 3.99–4.27 (*m*, 3H, C*H*CH_2, NCH_2), 6.29 (*d*, $J = 1.2$ Hz, 1H, CCHCO), 7.57–7.77 (*m*, 7H, H_{ar}), 8.01, 8.19 (2*s*, 0.6:0.4(1H), N=CH), 11.68, 11.76 (2*s*, 0.6:0.4(1H), NH) ppm; ^{13}C NMR (101 MHz, DMSO-d_6): δ 18.5 (CH_3), 33.1 (*C*H_2CO), 35.1 (*C*HCO), 50.5 (NCH_2), 106.3, 113.3, 115.4, 123.9, 126.2, 129.4, 132.3, 133.9, 142.6, 143.1, 146.4, 153.4, 153.9, 160.4 (C_{ar}, *CC*HCO, N=CH), 169.1, 173.5, 173.9 (3C=O) ppm. Anal. Calcd. for $C_{22}H_{18}BrN_3O_4$: C, 56.42; H, 3.87; N, 8.97 %. Found: C, 56.52; H, 3.68; N, 9.08 %.

*1-(4-Methyl-2-oxo-2H-chromen-7-yl)-5-oxo-N′-(propan-2-ylidene)pyrrolidine-3-carbohydrazide (**20**)*

A mixture of hydrazide **4** (0.30 g, 1 mmol) and acetone (20 mL) was heated at reflux for 3 h. The liquid fractions were evaporated under reduced pressure, and the residue was diluted with diethyl ether (10 mL). The formed precipitate was filtered off, washed with diethyl ether, and recrystallized from acetone to afford white solid, yield 0.23 g (67 %), mp 258–259 °C; IR (KBr): 3050 (NH), 1721, 1697, 1666 (3C=O), 1647 (C=N) cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 1.87, 1.89, 1.93, 1.94 (4*s*, 6H, $NC(CH_3)_2$), 2.40 (*s*, 3H, CH_3), 2.66–2.89 (*m*, 2H, CH_2CO), 3.54–3.37 (*m*, 1H, C*H*CH_2), 3.89–4.20 (*m*, 2H, NCH_2), 6.28 (*d*, $J = 1.2$ Hz, 1H, CCHCO), 7.65–7.81 (*m*, 3H, H_{ar}), 10.29, 10.37 (2*s*, 0.5:0.5(1H), NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ 17.1, 17.6, 17.9 ($3CH_3$), 24.9, 25.2 (*C*H_2CO), 35.2 (*C*HCO), 50.1, 50.8 (NCH_2), 105.6, 105.7, 112.7, 114.7, 125.7, 142.1, 151.4, 152.4, 153.3, 156.3, 159.9 (C_{ar}, *CC*HCO, N=C), 168.4, 173.1, 173.4 (3C=O) ppm. Anal. Calcd. for $C_{18}H_{19}N_3O_4$: C, 63.33; H, 5.61; N, 12.31 %. Found: C, 63.25; H, 5.50; N, 12.25 %.

*N-(2,5-Dimethyl-1H-pyrrol-1-yl)-1-(4-methyl-2-oxo-2H-chromen-7-yl)-5-oxopyrrolidine-3-carboxamide (**21**)*

A mixture of hydrazide **4** (0.30 g, 1 mmol), 2,5-hexanedione (0.14 g, 1.2 mmol), propan-2-ol (15 mL), and glacial acetic acid (1 mL) was heated at reflux for 5 h, cooled to room temperature, and diluted with water (20 mL). The formed precipitate was filtered off, washed with water, dried, and recrystallized from propan-2-ol and water mixture (2:1) to afford white solid, yield 0.26 g (69 %), mp 282–283 °C; IR (KBr): 3269 (NH), 1723, 1691, 1668 (3C=O) cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 2.00 (*s*, 6H, $2NCCH_3$), 2.41 (*d*, $J = 1.1$ Hz, 3H, CH_3), 2.74–3.04 (*m*, 2H, CH_2CO), 3.45–3.56 (*m*, 1H, C*H*CH_2), 4.00–4.27 (*m*, 2H, NCH_2), 5.65 (*s*, 2H, 2CH), 6.30 (*d*, $J = 1.2$ Hz, 1H, CCHCO), 7.70–7.79 (*m*, 3H, H_{ar}), 10.96 (*s*, 1H, NH) ppm. Anal. Calcd. for $C_{21}H_{21}N_3O_4$: C, 66.48; H, 5.58; N, 11.08 %. Found: C, 66.39; H, 5.39; N, 11.15 %.

*4-[(3,5-Dimethyl-1H-pyrazol-1-yl)carbonyl]-1-(4-methyl-2-oxo-2H-chromen-7-yl)pyrrolidin-2-one (**22**)*

A mixture of hydrazide **4** (0.30 g, 1 mmol), 2,4-pentanedione (1 g, 1.2 mmol), propan-2-ol (20 mL), and hydrochloric acid (0.5 mL) was heated at reflux for 5 h. Then it was cooled to room temperature, the precipitate was filtered off, washed with propan-2-ol, and recrystallized from propan-2-ol to afford white solid, yield 0.25 g (68 %), mp 229–230 °C; IR (KBr): 1727, 1715, 1699 (3C=O), 1584 (C=N) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 2.21 (*s*, 3H, N=CCH_3), 2.40 (*s*, 3H, CH_3), 2.48 (*s*, 3H, N–CCH_3), 2.85–3.04 (*m*, 2H, CH_2CO), 4.07–4.30 (*m*, 2H, NCH_2), 4.44–4.57 (*m*, 1H, C*H*CH_2), 6.24 (*s*, 1H, CH_{pyr}), 6.29 (*s*, 1H, CCHCO), 7.64–7.77 (*m*, 3H, H_{ar}) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ 13.5, 13.9, 17.9 ($3CH_3$), 35.1 (*C*H_2CO), 35.2 (*C*HCO), 50.0 (NCH_2), 105.8, 111.6, 112.7, 114.8, 115.5, 125.6, 141.9, 143.8, 152.1, 152.8, 153.3, (C_{ar}, *CC*HCO, C_{pyr}), 159.8, 172.3, 172.4 (3C=O) ppm. Anal. Calcd. for $C_{20}H_{19}N_3O_4$: C, 65.74; H, 5.24; N, 11.50 %. Found: C, 65.87; H, 5.38; N, 11.36 %.

*4-(5,6-Diphenyl-1,2,4-triazin-3-yl)-1-(4-methyl-2-oxo-2H-chromen-7-yl)pyrrolidin-2-one (**23**)*

A mixture of hydrazide **4** (0.60 g, 2 mmol), 1,2-diphenyl-1,2-ethanedione (0.42 g, 2 mmol), ammonium acetate (1.54 g, 20 mmol), and glacial acetic acid (30 mL) was heated at reflux for 24 h. Then it was cooled to room temperature and dissolved in water (30 mL). The precipitate was filtered off, washed with water, dried, and purified by column chromatography (methanol:chloroform, 1:5), $R_f = 0.48$ to afford white solid, yield 0.49 g (52 %), mp 269–270 °C; IR (KBr): 1720, 1698 (2C=O), 1589 (C=N) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 2.42 (*d*, $J = 1.2$ Hz, 3H, CH_3), 2.64–2.84 (*m*, 2H, CH_2CO), 3.19–3.20 (*m*, 1HC*H*CH_2), 3.90–4.11 (*m*, 2H, NCH_2), 6.31 (*d*, $J = 1.2$ Hz, 1H, CCHCO), 7.13–8.00 (*m*, 13H, H_{ar}) ppm. Anal. Calcd. for $C_{29}H_{22}N_4O_3$: C, 73.40; H, 4.67; N, 11.81 %. Found: C, 73.20; H, 4.75; N, 11.74 %.

*1-(4-Methyl-2-oxo-2H-chromen-7-yl)-4-(1,3,4-oxadiazol-2-yl)pyrrolidin-2-one (**24**)*

A mixture of hydrazide **4** (0.60 g, 2 mmol), triethoxymethane (1.06 g, 10 mmol), and 4-methylbenzenesulfonic acid (0.1 g, 0.53 mmol) was heated at reflux for 8 h. Then it was cooled to room temperature. The precipitate was filtered off, washed with water, dried, and recrystallized from ethanol to afford white solid, yield 0.35 g (56 %), mp 186–187 °C; IR (KBr): 1712, 1690 (2C=O), 1582, 1553 (2C=N) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 2.42 (*d*, $J = 1.2$ Hz 3H, CH_3), 2.91–3.19 (*m*, 2H, CH_2CO), 4.11–4.43 (*m*, 3H, C*H*CH_2 , and NCH_2), 6.32 (*s*, 1H, CCHCO), 7.65–7.82 (*m*, 3H, H_{ar}), 9.26 (*s*, 1H, OCHN) ppm. Anal. Calcd. for $C_{16}H_{13}N_3O_4$: C, 61.73; H, 4.21; N, 13.50 %. Found: C, 61.59; H, 4.33; N, 13.65 %.

*1-(4-Methyl-2-oxo-2H-chromen-7-yl)-4-(5-thioxo-4,5-dihydro-1,3,4-oxadiazol-2-yl)pyrrolidin-2-one (**25**)*

A mixture of hydrazide **4** (0.60 g, 2 mmol), potassium hydroxide (0.45 g, 8 mmol), carbon disulfide (0.38 g, 5 mmol), and methanol (20 mL) was heated at reflux for 24 h. The liquid fractions were evaporated under reduced pressure. The obtained residue was dissolved in water (20 mL), and the solution was acidified with hydrochloric acid to pH 2. The formed precipitate was filtered off, washed with water, and dried to afford light brown solid, yield 0.48 g (70 %), mp 252–253 °C; IR (KBr): 3055 (NH), 1715, 1687 (2C=O), 1613 (C=N), 1173 (C=S) cm^{-1}; ^1H NMR (700 MHz, DMSO-d_6): δ 2.41 (*d*, $J = 1.2$ Hz, 3H, CH_3), 2.89–3.10 (*m*, 2H, CH_2CO), 3.97–4.04 (*m*, 1H, C*H*CH_2), 4.14–4.39 (*m*, 2H, NCH_2), 6.30 (*d*, $J = 1.2$ Hz, 1H, CCHCO), 7.66–7.79 (*m*, 3H, H_{ar}), 14.46 (*s*, 1H, NH) ppm; ^{13}C NMR (176 MHz, DMSO-d_6): δ 18.0 (CH_3), 27.8 (*C*H_2CO), 35.3 (*C*HCO), 49.8 (NCH_2), 106.1, 112.9, 115.1, 115.6, 125.7, 141.8, 152.9, 153.4, (C_{ar}, *CC*HCO), 159.9, 171.9 (2C=O), 163.7 (C=N), 178.0 (C=S) ppm. Anal. Calcd. for $C_{16}H_{13}N_3O_4S$: C, 55.97; H, 3.82; N, 12.24 %. Found: C, 55.69; H, 3.91; N, 12.08 %.

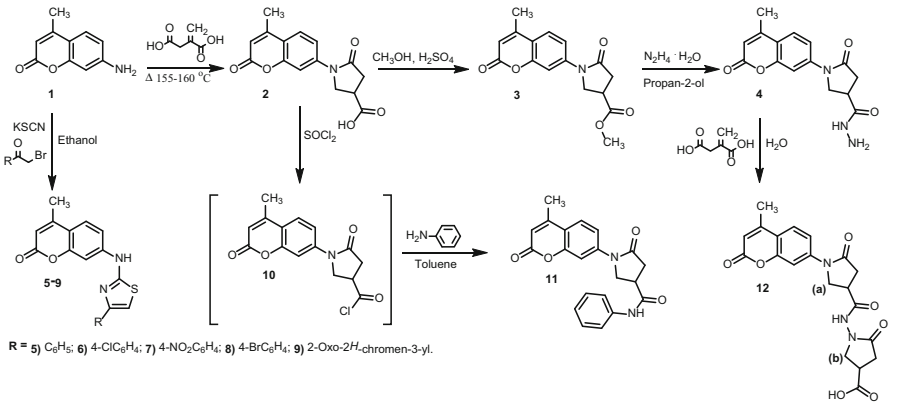
Biology

Antibacterial activity was tested by disk diffusion technique [42]. Microbial agents *Rhizobium radiobacter*, *Escherichia coli*, *Xanthomonas campestris* were commercially available from the German Collection of Microorganisms and Cell Cultures (DSMZ). The zone of inhibition of bacterial growth was investigated. The main solutions (500–1000 μg/cm^3) of the synthesized compounds were prepared in DMSO. Cultures of *R. radiobacter*, *E. coli*, and *X. campestris* were cultivated in Petri dishes on Luria–Bertani (LB) agar medium at 37 °C for 24 h. A bacterial suspension was prepared from cultivated bacterial cultures and 50 μL inoculum containing bacterial cells (108 CFU/cm^3) was spread over the LB agar medium. Filter paper disks were prepared by adding 25 μL of each compound solution and then disks were placed on the LB agar medium. Ampicillin was used as the positive control. The Petri dishes were incubated at 37 °C for 24 h and zones of inhibition were then measured for each sample.

Results and discussion

Chemistry

It is known that aromatic amines react with itaconic acid in water to form amino acids, which in the course of the reaction cyclize into 5-oxopyrrolidine-3-carboxylic acids [43, 44]. However, under these conditions 7-amino-4-methyl-2*H*-chromen-2-one (**1**) did not react with itaconic acid. 1-(4-Methyl-2-oxo-2*H*-chromen-7-yl)-5-oxopyrrolidine-3-carboxylic acid (**2**) was obtained by melting amine **1** with itaconic acid at 155–160 °C (Scheme 1). The structure of **2** has been confirmed by the presence of the ^1H NMR resonances of methylene group protons in pyrrolidinone



Scheme 1 Synthesis of *N*-substituted 7-amino-4-methyl-2*H*-chromen-2-ones **2–12**

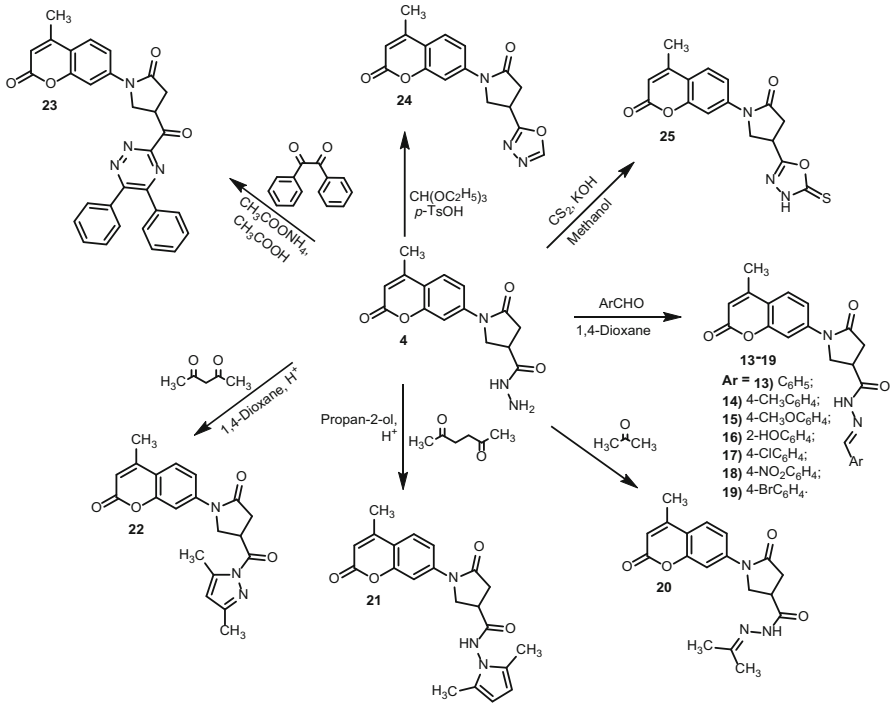
ring—a multiplet attributable to the $COCH_2$ group in the range of 2.49–2.53 ppm, a more deshielded multiplet of NCH_2 group in the range of 3.99–4.17 ppm, as well as a multiplet attributable to the CH group in the 3.28–3.46 ppm region. Three carbon lines at 159.8, 172.6, 173.9 ppm characteristic of the carbonyl groups and signals of pyrrolidinone ring carbon atoms at 34.8, 35.3, and 49.8 ppm attributable to CH_2 and CH groups are present in the ^{13}C NMR spectrum. Absorption lines of three C=O groups are seen at 1729, 1720, and 1671 cm^{-1} in the IR spectrum for **2**.

Acid hydrazides are more easily obtained from esters than acids; therefore, esterification of pyrrolidine-3-carboxylic acid **2** with methanol in the presence of sulfuric acid as a catalyst was conducted. The resulting methyl ester **3** was further treated with the excess of hydrazine hydrate in propan-2-ol at the reflux temperature to provide 1-(4-methyl-2-oxo-2*H*-chromen-7-yl)-5-oxopyrrolidine-3-carbohydrazide (**4**). A singlet of ester methoxy group protons at 3.69 ppm is absent in the ^1H NMR spectrum of hydrazide **4**; furthermore, a singlet attributable to the NH proton at 9.31 ppm and a broad signal of amine group protons at 4.35 ppm are observed thus confirming the structure of hydrazide. Absorption lines characteristic of the NH and NH_2 groups are present at 3267 and 3107 cm^{-1} in its IR spectrum. A modified *Hantzsch* method was applied to form thiazole ring in compounds **5–9** from amines and inorganic thiocyanates instead of thioamides and thiocarbamides. A corresponding α-haloketone was heated with potassium thiocyanate in ethanol at reflux temperature, and afterwards amine **1** was added into the reaction mixture, which was subsequently heated at reflux for approx. 40 h to give thiazole derivatives **5–9** in 54–68 % yield. In order to synthesize 5-oxopyrrolidine-3-carboxylic acid amide **11**, a mixture of pyrrolidine-3-carboxylic acid **2** and thionyl dichloride was heated at reflux to provide carboxylic acid chloride **10**, which, without isolation from the reaction mixture, was treated with aniline in toluene at the reflux temperature of the reaction mixture. In the ^1H NMR spectrum of **11**, additional resonances attributable to protons of benzene ring at the 7–8 ppm region and the NH group proton signal at 10.27 ppm are observed in comparison with the spectrum of **2**.

During the reaction of hydrazide **4** with itaconic acid, hydrazide, like primary amines, initially forms a compound containing γ-amino acid fragment, which in the course of the reaction cyclizes into a pyrrolidone ring. Thus, synthesized compound **12** contains two pyrrolidone rings linked by an amide bond and a chromenone ring. An analogous compound structure with aromatic substituents is described in the work [45]. Compound **12** contains two asymmetric carbon atoms; therefore, isomers of different configurations *R,R* and *S,S* or *R,S* and *S,R* are possible. The actual arrangement of substituents around these carbon atoms was not investigated because the attempts to obtain crystals suitable for X-ray analysis failed.

Hydrazones are characterised by broad biological activity spectrum among which numerous derivatives containing antispasmodic, antidepressant, anti-inflammatory, pain relieving, antithrombotic, antimicrobial, antiviral, and antitumor activities have been discovered [46].

Condensation of 1-(4-methyl-2-oxo-2*H*-chromen-7-yl)-5-oxopyrrolidine-3-carbohydrazide (**4**) with aromatic aldehydes and acetone was carried out to obtain hydrazones **13**–**20** (Scheme 2). A singlet attributable to the amine group is absent in their ^{1}H NMR spectra compared to the spectrum of **4**. Additionally, signals characteristic of aromatic protons occur in the range of 7–8 ppm. Because of the restricted rotation around amide bond, hydrazones are able to form *E/Z* isomers. Protons of NH and N=CH groups are observed in two line sets at 11.68 and



Scheme 2 Synthesis of *N*-substituted 7-amino-4-methyl-2*H*-chromen-2-ones **13**–**25**

11.76 ppm (NH), and 8.01 and 8.19 ppm (N=CH) in the ^{1}H NMR spectrum of compound **19**. According to the integration curve and the literature data [47] claiming that the line corresponding to a *Z* isomer is observed in the stronger magnetic field due to the stronger shielding, it can be stated that *Z* isomer dominates 60 % of the *E/Z* isomer mixture of compound **19**. An identical ratio of *E/Z* isomers in the DMSO-d_6 solutions has been determined from the spectra of the other synthesized hydrazones.

Condensation of 5-oxopyrrolidine-3-carbohydrazide **4** with 2,5-hexanedione in propan-2-ol at the reflux temperature in the presence of acetic acid as a catalyst furnished *N*-substituted pyrrole derivative **21**. In its ^{1}H NMR spectrum of this compound, a singlet corresponding to the protons of two CH_3 groups in pyrrole ring is observed at 2.00 ppm as well as a singlet of two =CH group protons at 5.65 ppm, whereas an analogous reaction of hydrazide **4** with 2,4-pentandione in the presence of hydrochloric acid resulted in formation of 4-(3,5-dimethylpyrazol-1-carbonyl)-1-(4-methyl- 2-oxo-2*H*-chromen-7-yl)pyrrolidin-2-one (**22**). Its structure has been confirmed by the presence of two ^{1}H NMR resonances attributable to two CH_3 group protons at 2.21 and 2.48 ppm, and a singlet of the CH group proton in pyrazole ring at 6.24 ppm. Carbon resonances ascribed to two CH_3 group carbon atoms are observed at 13.5 and 13.9 ppm in the ^{13}C NMR spectrum.

The reaction of hydrazide **4** with 1,2-diphenyl-1,2-ethanedione in acetic acid at the reflux temperature did not provide an expected hydrazone type non-cyclic compound. However, when ammonium acetate was added to the reaction mixture, compound **23** containing 1,2,4-triazine cycle was obtained during the ternary reaction. An increased number of the protons resonating in the aromatic region of the ^{1}H NMR spectrum along with the data of IR spectroscopy and elemental analysis have proven the structure of **23**.

Oxadiazoles are an important class of heterocyclic compounds characterized by pharmacological activity [48]. 1-(4-Methyl-2-oxo-2*H*-chromen-7-yl)-4-(1,3,4-oxadiazole-2-yl)pyrrolidin-2-one (**24**) was synthesized by heating at reflux hydrazide **4** with an excess of triethoxymethane in the presence of 4-methylbenzenesulfonic acid as a catalyst. A singlet attributable to the CH group proton in oxadiazole ring is observed at 9.26 ppm in the ^{1}H NMR spectrum for **24**. In the IR spectrum, the absorption lines characteristic of carbonyl groups are observed at 1712 and 1690 cm^{-1}, the ones corresponding to the C=N group in oxadiazole ring are at 1582 and 1553 cm^{-1}. 1,3,4-Oxadiazole **25** was prepared by heating at reflux a mixture of hydrazide **4**, carbon disulfide, and potassium hydroxide in methanol, followed by the dissolution of the resulting potassium dithiocarbazate in water and treatment of the obtained solution with hydrochloric acid to pH 2.

Biology

Some of the synthesized compounds (**4**–**9**, **11**–**25**) were screened for their antibacterial activity against *R. radiobacter*, *E. coli*, and *X. campestris*. Strains of the *Rhizobium* species (formerly *Agrobacterium*, which was reclassified based on 16S rDNA analyses) are aerobic, motile, oxidase-positive, and non-spore-forming Gram-negative bacilli. Among the species of *Rhizobium* (i.e. *R. radiobacter, R.*

rhizogenes, R. rubi, R. undicola, and *R. vitis)*, *R. radiobacter* is the species that most commonly causes disease in humans. Since the first case of human infection with *R. radiobacter* in a patient with prosthetic aortic valve endocarditis was reported in 1980, *R. radiobacter* has been recognized as an opportunistic human pathogen. Most patients with *R. radiobacter* infection have debilitating underlying diseases [49]. Clinical manifestations include septicemia, bacteremia, prosthetic valve endocarditis, urinary tract infection, peritonitis, and pneumonia [50]. Bacteria belonging to the genus *Xanthomonas* are one of the most omnipresent groups of Gram-negative plant pathogenic bacteria and cause a variety of diseases in multiple plants [51]. *X. campestris* pv. *campestris* (Xcc), the cause of black rot in crucifers, is a seed-borne bacterium that occurs worldwide [52].

Table 1 Antibacterial activity of compounds **4–9** and **11–25** against *Rhizobium radiobacter, Escherichia coli, Xanthomonas campestris*

Compound	*R. radiobacter*		*E. coli*		*X. campestris*	
	750 μg/cm^3	1000 μg/cm^3	750 μg/cm^3	1000 μg/cm^3	750 μg/cm^3	1000 μg/cm^3
4	–	+	–	–	–	+
5	–	+	+	+	+	+
6	–	–	–	+	–	+
7	–	+	–	+	–	+
8	–	–	+	+	+	+
9	–	+	+	+	–	+
11	–	–	–	–	–	–
12	–	–	–	–	–	–
13	+	+	–	–	–	+
14	+	+	–	–	–	–
15	–	+	–	–	–	–
16	–	–	–	+	–	+
17	–	–	–	+	–	–
18	–	+	–	+	–	–
19	–	–	–	+	–	–
20	–	–	–	–	–	–
21	–	–	–	–	–	–
22	–	–	–	–	–	–
23	–	+	–	–	–	+
24	–	–	–	+	–	–
25	–	+	–	–	+	+
Ampicillin (50 μg/ml)	23*		6.4		19.7	

\+ The compound exhibited antibacterial activity

– The compound did not exhibit antibacterial activity

* Values mm

Antibacterial activities of the compounds were tested by the diffusion technique at 500, 750, and 1000 μg/cm^3 concentrations and were compared with that of the known antibacterial agent ampicillin. As seen from the data presented in Table 1, the tested compounds showed a weak antibacterial activity. At 750 μg/cm^3 concentration *R. radiobacter* was sensitive to compounds **13** and **14**. At the same concentration, compounds **5**, **8**, and **9** were active against *E. coli*, whereas compounds **5**, **8**, and **25** exhibited antibacterial activity against *X. campestris*. None of the synthesized compounds exhibited antibacterial activity at 500 μg/cm^3.

Conclusions

In summary, various *N*-substituted 7-amino-4-methyl-2*H*-chromen-2-ones with functionalized azole, diazole, oxazole, and thiazole substituents were synthesized. The screening of the antibacterial activity of the synthesized compounds has revealed that hydrazones **13** and **14** are the most active against *R. Radiobacter* (750 μg/cm^3), *E. coli* was the most sensitive to the derivatives of 4-methyl-7-[(4-*R*-1,3-thiazol-2-yl)amino]-2*H*-chromen-2-one containing 1,3-thiazole moiety **5**, **8**, and **9** (750 μg/cm^3), and *X. campestris* was the most sensitive to 1,3-thiazoles **5** and **8** as well as 5-thioxo-1,3,4-oxadiazole **25** (750 μg/cm^3).

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