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MEDICINAL CHEMISTRY RESEARCH

ORIGINAL RESEARCH

Synthesis of coumarin-based 1,3,4-oxadiazol-2ylthio-*N*-phenyl/benzothiazolyl acetamides as antimicrobial and antituberculosis agents

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Abstract In an attempt to find new agents to fight against microbial infections, a series of coumarin-based 1,3,4-oxadiazol-2ylthio-*N*-phenyl/benzothiazolyl acetamides was synthesized starting from coumarin-3-carboxylic acid ethyl ester obtained through Knoevenagel and Pinner reaction. In vitro antimicrobial activity against several bacteria (*S. aureus*, *B. cereus*, *E. coli*, *P. aeruginosa*, *K. pneumoniae*, *S. typhi*, *P. vulgaris*, *S. flexneri*), fungi (*A. niger*, *A. fumigatus*, *A. clavatus*, *C. albicans*) and antimycobacterial activity against *Mycobacterium tuberculosis* H37Rv strain was assessed. This study shows to what extent the presence of various electron withdrawing/donating substituents on the phenyl or benzothiazole ring affects the activity profiles of the newer molecules. The relationship between activity profiles (MICs, 3.12–25 μg/mL) and the lipophilic character (Log*P*) of the prepared products is also discussed and the MIC values of the active conjugates seem to correlate to some extent with the lipophilicity profiles. Two (**5e** and **6c**) of the final analogues displayed promising antimycobacterial activity at 12.5 μg/mL of MIC, half fold potent to the standard drug pyrazinamide (6.25 μg/mL). Compounds were characterized by IR, ^{1}H NMR, ^{13}C NMR spectroscopy and elemental analysis.

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Introduction

During the past two decades, the growing population of immunocompromised individuals, patients with malignancies and transplant recipients has resulted in an increase in severe opportunistic microbial infections (Nathan, 2004). On the other hand, tuberculosis (TB), a disease long considered substantially eradicated in the developed countries, has resurged dramatically in the last decades, establishing itself as one of the infectious diseases resulting in around two million human deaths worldwide (Raviglione, 2003; NIAID, http://www3.niaid.nih.gov/topics/tuberculosis/; TAACF, http://www.taacf.org/about-TB-background.htm). According to the World Health Organization (WHO), in 2010, there were 8.8 million (range 8.5–9.2 million) incident cases of TB, 1.1 million (range 0.9–1.2 million) deaths from TB among HIV-negative people and an additional 0.35 million (range 0.32–0.39 million) deaths from HIV-associated TB (WHO, 2011). These health problems demand urgency to develop new authentically genuinely broad-spectrum and low-toxicity antimicrobial and antitubercular drugs as strains that resist the current medications are spreading across the globe quite fast.

Coumarin and its derivatives have gained great therapeutic importance in the field of medicinal chemistry since they display a fascinating array of pharmacological properties (Keating and O'Kennedy, 1997; Li *et al*., 2011; Hishmat *et al*., 2008; De Souza *et al*., 2005; Yee *et al*., 2005). Moreover, it has been reported in the literature that

compounds bearing coumarin-based 1,3,4-oxadiazole ring is well acknowledged to possess significant antimicrobial activity (Bhat *et al*., 2011; Patel *et al*., 2010; Biradar and Manjunath, 2004; Desai and Mehta, 2002). Looking at the pharmacological importance of above-mentioned scaffolds and in continuous interest in oxadiazole derivatives as antimicrobial agents (Chikhalia *et al*., 2009), we were interested in identifying newer structural elements consisting of both coumarin and oxadiazole nucleus to unearth novel antibacterial, antifungal and antimycobacterial agents. Furthermore, various analogues carrying benzothiazole ring system have been proved to be effective to possess promising antituberculosis activities (Bhusari *et al*., 2000), whereas imidazo[2,1-*b*][1,3,4]thiadiazoles-based benzothiazole derivatives are evaluated for their in vitro antituberculosis activity (Hegde *et al*., 2006) using BACTEC method and the analogues were found to reveal excellent inhibition (88–99 %) of mycobacteria at 6.25 μg/mL of MIC. Moreover, recent literature is enriched with progressive findings about the different biological activities of benzothiazole derivatives (Rana *et al*., 2007).

We have previously developed some 1,3,5-triazine-based analogues bearing coumarin nucleus and identified their antimicrobial efficacies (Patel *et al*., 2011a) and the noticeable results directed us to assess the antimicrobial potency of some additional 1,3,5-triazine-based derivatives bearing 3-(5-mercapto-1,3,4-oxadiazol-2-yl)-2*H*-chromen-2-one (Patel *et al*., 2011b) and results suggested that such coumarin-based 1,3,4-oxadiazolyl derivatives are important starting tool for the antimicrobial drug discovery process. These findings have given impetus to our antimicrobial drug research and we thought that structural modification of the existed molecules to a different way involving coumarin and 1,3,4-oxadiazole core could modulate the antimicrobial profiles in a more better way. Furthermore, the physicochemical parameter like Log*P* refers to calculated hydrophobicity of the compounds and according to the thumb rule for Log*P* values, to a drug like molecule it must be lower than "5" to by-pass the cell barrier. To describe the uptake, distribution, biotransformation and excretion of organic chemicals in biological systems, partition or distribution coefficient is critical elements (Jepson *et al*., 1993) and hence, in this context, the newer molecules with appreciable lipophilic character are presented here to produce remarkable bioactivities.

Materials and methods

Melting points were determined in open capillaries on a Veego electronic apparatus VMP-D (Veego Instrument Corporation, Mumbai, India) and are uncorrected. IR spectra (4,000–400 cm^{-1}) of synthesized compounds were recorded on a Shimadzu 8400-S FT–IR spectrophotometer (Shimadzu India Pvt. Ltd., Mumbai, India) using KBr pellets. Thin layer chromatography was performed on object glass slides (2 × 7.5 cm) coated with silica gel-G and spots were visualized under UV irradiation. ^{1}H NMR and ^{13}C NMR spectra were recorded on a Varian 400 MHz model spectrometer (Varian India Pvt. Ltd., Mumbai, India) using DMSO as a solvent and TMS as internal standard with ^{1}H resonant frequency of 400 MHz and ^{13}C resonant frequency of 100 MHz. The ^{1}H NMR and ^{13}C NMR chemical shifts were reported as parts per million (ppm) downfield from TMS (Me_4Si) and were performed at Centre for Excellence, Vapi, India. The splitting patterns are designated as follows: s singlet, br s broad singlet, d doublet, m multiplet. Elemental analyses (C, H, N) were performed using a Heraeus Carlo Erba 1180 CHN analyser (Hanau, Germany).

General synthetic procedure for 2-chloro-*N*-(substituted)phenyl acetamides (**2a**–**k**)

Chloroacetyl chloride (0.06 mol) was added dropwise to a mixture of the appropriate amine (0.05 mol) and K_2CO_3 (0.06 mol) in acetone (50 mL) at room temperature. The reaction mixture was refluxed for 4–8 h, then, after cooling to room temperature, it was slowly poured into 100 mL of ice water. A solid was formed thereafter. The precipitate was separated by filtration and washed successively with water. The product was dried under vacuum to obtain **2a**–**k**. The progress of the reaction was monitored by thin layer chromatography using toluene–acetone (8:2) solvent system.

*2-Chloro-N-phenyl acetamide (**2a**)*

White solid, yield: 78 %., mp 135–138 °C. IR (KBr, cm^{-1}): 3,281 (NH), 1,680 (C=O). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.56 (br s, 1H, –NH), 7.66 (m, 2H), 7.40 (m, 2H), 7.14 (m, 1H), 4.22 (s, 2H, $–CH_2–Cl$).

*2-Chloro-N-(4-chlorophenyl)acetamide (**2b**)*

Pale yellow solid, yield: 83 %, mp 168–170 °C. IR (KBr, cm^{-1}): 3,292 (NH), 1,672 (C=O). ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.35 (br s, 1H, –NH), 7.63 (m, 2H), 7.45 (m, 2H), 7.19 (m, 1H), 4.21 (s, 2H, $–CH_2–Cl$).

*N-(4-Bromophenyl)-2-chloroacetamide (**2c**)*

Grey solid, yield: 67 %, mp 180–184 °C. IR (KBr, cm^{-1}): 3,307 (NH), 1,668 (C=O). ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.32 (br s, 1H, –NH), 7.54–7.47 (m, 3H), 7.22 (m, 1H), 4.25 (s, 2H, $–CH_2–Cl$).

*2-Chloro-N-(4-fluorophenyl)acetamide (**2d**)*

While solid, yield: 62 %, mp 131–132 °C. IR (KBr, cm^{-1}): 3,299 (NH), 1,670 (C=O). ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.21 (br s, 1H, –NH), 7.51 (dd, $J = 7.1$, 4.4 Hz, 2H), 7.27 (t, $J = 8.2$ Hz, 2H), 4.12 (s, 2H, $–CH_2–Cl$).

*2-Chloro-N-(4-iodophenyl)acetamide (**2e**)*

While solid, yield: 85 %, mp 192–196 °C. IR (KBr, cm^{-1}): 3,314 (NH), 1,678 (C=O). ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.44 (br s, 1H, –NH), 7.64 (d, $J = 7.1$ Hz, 2H), 7.42 (d, $J = 7.4$ Hz, 2H), 4.20 (s, 2H, $–CH_2–Cl$).

*2-Chloro-N-(4-nitrophenyl)acetamide (**2f**)*

Yellow solid, yield: 75 %, mp 185–186 °C. IR (KBr, cm^{-1}): 3,283 (NH), 1,669 (C=O). ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.41 (br s, 1H, –NH), 7.53–7.41 (m, 4H), 4.18 (s, 2H, $–CH_2–Cl$).

*2-Chloro-N-(4-cyanophenyl)acetamide (**2g**)*

While solid, yield: 88 %, mp 182–184 °C. IR (KBr, cm^{-1}): 3,270 (NH), 1,669 (C=O). ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.32 (br s, 1H, –NH), 7.67 (d, $J = 7.3$ Hz, 2H), 7.58 (d, $J = 7.4$ Hz, 2H), 4.14 (s, 2H, $–CH_2–Cl$).

*2-Chloro-N-p-tolylacetamide (**2h**)*

While solid, yield: 80 %, mp 164–166 °C. IR (KBr, cm^{-1}): 3,295 (NH), 1,671 (C=O). ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.30 (br s, 1H, –NH), 7.52–7.49 (m, 2H), 7.34–7.36 (m, 2H), 4.21 (s, 2H, $–CH_2–Cl$), 2.19 (s, 3H, $–CH_3$).

*2-Chloro-N-(4-methoxyphenyl)acetamide (**2i**)*

While solid, yield: 76 %, mp 121–122 °C. IR (KBr, cm^{-1}): 3,311 (NH), 1,668 (C=O). ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.27 (br s, 1H, –NH), 7.60 (d, $J = 7.5$ Hz, 2H), 7.44 (d, $J = 7.1$ Hz, 2H), 4.24 (s, 2H, $–CH_2–Cl$), 3.77 (s, 3H, $–OCH_3$).

*2-Chloro-N-(4-ethoxyphenyl)acetamide (**2j**)*

While solid, yield: 68 %, mp 189–193 °C. IR (KBr, cm^{-1}): 3,319 (NH), 1,664 (C=O). ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.39 (br s, 1H, –NH), 7.55 (d, $J = 7.3$ Hz, 2H), 7.38 (d, $J = 7.4$ Hz, 2H), 4.22 (s, 2H, $–CH_2–Cl$), 3.97 (q, $J = 5.9$ Hz, 2H, $–O–CH_2–$), 1.49 (t, $J = 5.8$ Hz, 3H, $–CH_3$).

*N-(4-Acetamidophenyl)-2-chloroacetamide (**2k**)*

Pale yellow solid, yield: 72 %, mp 240–243 °C. IR (KBr, cm^{-1}): 3,290 (NH), 1,671 (C=O). ^{1}H NMR (400 MHz, DMSO-d_6): δ 8.21 (br s, 1H, –NH), 8.33 (s, 1H, –NH), 7.63–7.56 (m, 2H), 7.39–7.35 (m, 2H), 4.20 (s, 2H, $–CH_2–Cl$), 1.91 (s, 3H, $–CH_3$).

General synthetic procedure for 2-amino-6-substituted benzothiazoles (**3a**–**k**)

A mixture of 0.1 mol of 4-substituted aniline and 0.1 mol of potassium thiocyanate (KCNS) in 100 mL glacial acetic acid (AcOH) was cooled in an ice bath and stirred for 10–20 min, and then 0.1 mol bromine in glacial acetic acid was added dropwise at such a rate to keep the temperature below 10 °C throughout the addition. The reaction mixture was stirred at room temperature for 2–4 h, the hydrobromide (HBr) salt thus separated out was filtered, washed with acetic acid, dried, dissolved in hot water and basified to pH 11.0 with ammonia solution (NH_4OH) and the resulting precipitate was filtered, washed with water and dried to get the desired product (**3a**–**k**). The progress of the reaction was monitored by thin layer chromatography using toluene–acetone (8:2) solvent system.

*Benzo[d]thiazol-2-amine (**3a**)*

Light brown solid, yield: 78 %, mp 128–130 °C. IR (KBr, cm^{-1}): 3,371 (NH), 1,578 (C=N). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 7.28 (s, 2H, NH_2, D_2O exchangeable), 7.24–7.51 (m, 4H, Ar–H).

*6-Chlorobenzo[d]thiazol-2-amine (**3b**)*

White solid, yield: 72 %, mp 202–203 °C. IR (KBr, cm^{-1}): 3,391 (NH), 1,546 (C=N). ^{1}H NMR (400 MHz, DMSO-d_6): δ 7.52 (d, $J = 1.7$ Hz, 1H, H-7), 7.35 (d, $J = 7.3$ Hz, 1H, H-4), 7.21 (dd, $J = 7.9$, 2.1 Hz, 1H, H-5), 6.72 (s, 2H, NH_2, D_2O exchangeable).

*6-Bromobenzo[d]thiazol-2-amine (**3c**)*

Light yellow solid, yield: 76 %, mp 216–219 °C. IR (KBr, cm^{-1}): 3,410 (NH), 1,589 (C=N). ^{1}H NMR (400 MHz, DMSO-d_6): δ 7.72 (d, $J = 1.5$ Hz, 1H, H-7), 7.39–7.46 (m, 2H, H-4 and H-5), 6.72 (s, 2H, NH_2, D_2O exchangeable).

*6-Fluorobenzo[d]thiazol-2-amine (**3d**)*

Off while solid, yield: 80 %, mp 181–184 °C. IR (KBr, cm^{-1}): 3,368 (NH), 1,584 (C=N). ^{1}H NMR (400 MHz, DMSO-d_6): δ 7.59 (dd, $J = 2.4$, 8.3 Hz, 1H, H-7), 7.34

(dd, $J = 4.8$, 8.6 Hz, 1H, H-4), 7.16 (dt, $J = 2.2$, 8.8 Hz, 1H, H-5), 7.09 (s, 2H, NH_2, D_2O exchangeable).

*6-Iodobenzo[d]thiazol-2-amine (**3e**)*

Off while solid, yield: 70 %, mp 206–208 °C. IR (KBr, cm^{-1}): 3,290 (NH), 1,573 (C=N). ^1H NMR (400 MHz, DMSO-d_6): δ 7.79 (d, $J = 1.9$ Hz, 1H, H-7), 7.51 (d, $J = 7.1$ Hz, 1H, H-4), 7.16 (dd, $J = 8.1$, 2.2 Hz, 1H, H-5), 6.92 (s, 2H, NH_2, D_2O exchangeable).

*6-Nitrobenzo[d]thiazol-2-amine (**3f**)*

Yellow solid, yield: 74 %, mp 248–251 °C. IR (KBr, cm^{-1}): 3,402 (NH), 1,569 (C=N). ^1H NMR (400 MHz, DMSO-d_6): δ 7.62 (d, $J = 1.9$ Hz, 1H, H-7), 7.29–7.37 (m, 2H, H-4 and H-5), 6.22 (s, 2H, NH_2, D_2O exchangeable).

*2-Aminobenzo[d]thiazole-6-carbonitrile (**3g**)*

A mixture of 0.1 mol of 4-aminobenzonitrile and 0.2 mol of potassium thiocyanate (KSCN) in 100 mL glacial acetic acid was cooled in an ice bath and stirred for 10–20 min, and then 0.1 mol of bromine in glacial acetic acid was added dropwise at such a rate to keep the temperature below 10 °C throughout the addition. The reaction mixture was stirred at room temperature for 2–4 h, the HBr salt thus separated out. In the reaction mixture 150 mL water was added. The solid 4-cyano-2-thiocyanatoaniline thus obtained was filtered, dried and recrystallized from ethanol. Then in 0.05 mol of this intermediate, 10 mL of concentrated HCl and 20 mL of water was added and the mixture was refluxed for 2 h. The progress of the reaction was monitored by thin layer chromatography using toluene–acetone (9:1) solvent system. After the completion of the reaction, the solution was cooled and the product was filtered, washed with water and recrystallized from ethanol to give **3g** (Sawhney and Boykin, 1979). Light yellow solid, yield: 61 %, mp 206–207 °C. IR (KBr, cm^{-1}): 3,376 (NH), 1,576 (C=N). ^1H NMR (400 MHz, DMSO-d_6): δ ppm, 7.70 (d, $J = 1.8$ Hz, 1H, H-7), 7.55 (d, $J = 7.4$ Hz, 1H, H-4), 7.26 (dd, $J = 8.4$, 2.0 Hz, 1H, H-5), 7.18 (s, 2H, NH_2, D_2O exchangeable).

*6-Methylbenzo[d]thiazol-2-amine (**3h**)*

Light yellow solid, yield: 77 %, mp 144–145 °C. IR (KBr, cm^{-1}): 3,393 (NH), 1,570 (C=N). ^1H NMR (400 MHz, DMSO-d_6): δ 7.53–7.39 (m, 3H, H-4, H-5, H-7), 5.89 (s, 2H, NH_2, D_2O exchangeable), 1.96 (s, 3H, CH_3).

*6-Methoxybenzo[d]thiazol-2-amine (**3i**)*

Off white solid, yield: 71 %, mp 169–170 °C. IR (KBr, cm^{-1}): 3,278 (NH), 1,571 (C=N). ^1H NMR (400 MHz, DMSO-d_6): δ 7.62–7.53 (m, 2H, H-4, H-7), 7.20 (dd, $J = 8.2$, 1.9 Hz, 1H, H-5), 5.42 (s, 2H, NH_2, D_2O exchangeable), 3.80 (s, 3H, OCH_3).

*6-Ethoxybenzo[d]thiazol-2-amine (**3j**)*

Off white solid, yield: 74 %, mp 162–165 °C. IR (KBr, cm^{-1}): 3,390 (NH), 1,585 (C=N). ^1H NMR (400 MHz, DMSO-d_6): δ 7.53–7.37 (m, 2H, H-4, H-7), 7.27 (dd, $J = 8.6$, 1.6 Hz, 1H, H-5), 5.53 (s, 2H, NH_2, D_2O exchangeable), 4.08 (q, $J = 6.2$ Hz, 2H, OCH_2CH_3), 2.21 (t, $J = 6.4$ Hz, 3H, CH_2CH_3).

*N-(2-Aminobenzo[d]thiazol-6-yl)acetamide (**3k**)*

While solid, yield: 61 %, mp 141–143 °C. IR (KBr, cm^{-1}): 3,401 (NH), 1,573 (C=N). ^1H NMR (400 MHz, DMSO-d_6): δ 8.13 (s, 1H, –NH), 7.67–7.54 (m, 2H, H-4, H-7), 7.30 (dd, $J = 8.6$, 1.6 Hz, 1H, H-5), 7.15 (s, 2H, NH_2, D_2O exchangeable), 2.04 (s, 3H, CH_3).

General synthetic procedure for *N*-(benzo[d]thiazol-2-yl)-2-chloroacetamides (**4a–k**)

Chloroacetyl chloride (0.06 mol) was added dropwise to a mixture of the appropriate 2-amino-6-substituted benzothiazole (**3a–k**) (0.05 mol) and K_2CO_3 (0.06 mol) in benzene (50 mL) at room temperature. The reaction mixture was refluxed for 6–12 h, then, after cooling to room temperature, it was slowly poured into 100 mL of ice water. A solid was formed thereafter. The precipitate was separated by filtration and washed successively with water. The product was dried under vacuum to obtain **4a–k**. The progress of the reaction was monitored by thin layer chromatography using toluene–acetone (8:2) solvent system.

*N-(Benzo[d]thiazol-2-yl)-2-chloroacetamide (**4a**)*

Light brown solid, yield: 73 %, mp 135–138 °C. IR (KBr, cm^{-1}): 3,283 (NH), 1,679 (CO). ^1H NMR (400 MHz, DMSO-d_6): δ ppm, 8.43 (s, 1H, –NH), 7.19–7.38 (m, 4H, Ar–H), 4.23 (s, 2H, CH2–Cl).

*2-Chloro-N-(6-chlorobenzo[d]thiazol-2-yl)acetamide (**4b**)*

While solid, yield: 88 %, mp 207–210 °C. IR (KBr, cm^{-1}): 3,290 (NH), 1,689 (CO). ^1H NMR (400 MHz, DMSO-d_6): δ 8.56 (s, 1H, –NH), 7.64 (d, $J = 1.6$ Hz, 1H, H-7), 7.45

(d, $J = 7.6$ Hz, 1H, H-4), 7.27 (dd, $J = 8.1$, 1.8 Hz, 1H, H-5), 4.17 (s, 2H, $–CH_2–Cl$).

*N-(6-Bromobenzo[d]thiazol-2-yl)-2-chloroacetamide (**4c**)*

Light yellow solid, yield: 72 %, mp 195–197 °C. IR (KBr, cm^{-1}): 3,310 (NH), 1,671 (CO). ^1H NMR (400 MHz, DMSO-d_6): δ 8.43 (s, 1H, –NH), 7.60 (d, $J = 1.7$ Hz, 1H, H-7), 7.35–7.43 (m, 2H, H-4 and H-5), 4.06 (s, 2H, $–CH_2–Cl$).

*2-Chloro-N-(6-fluorobenzo[d]thiazol-2-yl)acetamide (**4d**)*

Off while solid, yield: 60 %, mp 177–179 °C. IR (KBr, cm^{-1}): 3,277 (NH), 1,665 (CO). ^1H NMR (400 MHz, DMSO-d_6): δ 8.39 (s, 1H, –NH), 7.70 (dd, $J = 2.3$, 8.5 Hz, 1H, H-7), 7.38 (dd, $J = 4.5$, 8.2 Hz, 1H, H-4), 7.23 (dt, $J = 2.4$, 8.9 Hz, 1H, H-5), 4.25 (s, 2H, $–CH_2–Cl$).

*2-Chloro-N-(6-iodobenzo[d]thiazol-2-yl)acetamide (**4e**)*

Off while solid, yield: 79 %, mp 197–200 °C. IR (KBr, cm^{-1}): 3,256 (NH), 1,680 (CO). ^1H NMR (400 MHz, DMSO-d_6): δ 8.46 (s, 1H, –NH), 7.58 (d, $J = 2.2$ Hz, 1H, H-7), 7.45 (d, $J = 7.4$ Hz, 1H, H-4), 7.29 (dd, $J = 8.5$, 1.9 Hz, 1H, H-5), 4.16 (s, 2H, $–CH_2–Cl$).

*2-Chloro-N-(6-nitrobenzo[d]thiazol-2-yl)acetamide (**4f**)*

Yellow solid, yield: 78 %, mp 218–220 °C. IR (KBr, cm^{-1}): 3,278 (NH), 1,671 (CO). ^1H NMR (400 MHz, DMSO-d_6): δ 8.55 (s, 1H, –NH), 7.60 (d, $J = 1.8$ Hz, 1H, H-7), 7.35–7.46 (m, 2H, H-4 and H-5), 4.28 (s, 2H, $–CH_2–Cl$).

*2-Chloro-N-(6-cyanobenzo[d]thiazol-2-yl)acetamide (**4g**)*

Light yellow solid, yield: 69 %, mp 225–227 °C. IR (KBr, cm^{-1}): 3,298 (NH), 1,685 (CO). ^1H NMR (400 MHz, DMSO-d_6): δ 8.37 (s, 1H, NH), 7.55 (d, $J = 1.6$ Hz, 1H, H-7), 7.43 (d, $J = 7.7$ Hz, 1H, H-4), 7.18 (dd, $J = 8.6$, 2.1 Hz, 1H, H-5), 4.20 (s, 2H, $–CH_2–Cl$).

*2-Chloro-N-(6-methylbenzo[d]thiazol-2-yl)acetamide (**4h**)*

Light yellow solid, yield: 53 %, mp 175–177 °C. IR (KBr, cm^{-1}): 3,311 (NH), 1,673 (CO). ^1H NMR (400 MHz, DMSO-d_6): δ 8.45 (s, 1H, NH), 7.63–7.46 (m, 3H, H-4, H-5, H-7), 4.20 (s, 2H, $–CH_2–Cl$), 1.81 (s, 3H, CH_3).

*2-Chloro-N-(6-methoxybenzo[d]thiazol-2-yl)acetamide (**4i**)*

Off white solid, yield: 75 %, mp 169–172 °C. IR (KBr, cm^{-1}): 3,291 (NH), 1,667 (CO). ^1H NMR (400 MHz, DMSO-d_6): δ 8.55 (s, 1H, NH), 7.71–7.64 (m, 2H, H-4, H-7), 7.33 (dd, $J = 8.3$, 2.0 Hz, 1H, H-5), 4.24 (s, 2H, $–CH_2–Cl$), 3.80 (s, 3H, OCH_3).

*2-Chloro-N-(6-ethoxybenzo[d]thiazol-2-yl)acetamide (**4j**)*

Off white solid, yield: 84 %, mp 179–181 °C. IR (KBr, cm^{-1}): 3,287 (NH), 1,679 (CO). ^1H NMR (400 MHz, DMSO-d_6): δ 8.40 (s, 1H, NH), 7.60–7.47 (m, 2H, H-4, H-7), 7.34 (dd, $J = 8.5$, 1.8 Hz, 1H, H-5), 4.26 (s, 2H, $–CH_2–Cl$), 4.18 (q, $J = 5.8$ Hz, 2H, OCH_2CH_3), 2.19 (t, $J = 6.7$ Hz, 3H, CH_2CH_3).

*N-(6-Acetamidobenzo[d]thiazol-2-yl)-2-chloroacetamide (**4k**)*

White solid, yield: 52 %, mp 227–229 °C. IR (KBr, cm^{-1}): 3,277 (NH), 1,670 (CO). ^1H NMR (400 MHz, DMSO-d_6): δ 8.57 (s, 1H, NH), 8.33 (s, 1H, –NH), 7.62–7.51 (m, 2H, H-4, H-7), 7.27 (dd, $J = 8.4$, 1.9 Hz, 1H, H-5), 4.21 (s, 2H, $–CH_2–Cl$), 2.04 (s, 3H, CH_3).

*Synthesis of ethyl 2-oxo-2H-chromene-3-carboxylate (**2**)*

Salicylaldehyde (**1**) (10 g, 0.045 mol) and diethylmalonate (7.34 g, 0.045 mol) were dissolved in ethanol (150 mL) to give clear solution. Piperidine (18 mL) was added and the mixture was refluxed for 5 h. After the completion of the reaction (monitored by thin layer chromatography in toluene–ethyl acetate solvent system), the content was concentrated to small volume. Then the reaction mixture was poured onto crushed ice and the resulted solid was filtered, dried and recrystallized from ethanol to afford (**3**) as a white solid, yield: 86 %, mp 124–125 °C. IR (KBr, cm^{-1}): 1,745 (C=O, ester), 1,725 (CO, coumarin), 1,677 (C=O), 1,231 (C–O). ^1H NMR (400 MHz, DMSO-d_6): δ ppm, 8.27 (1H, s, H-4, coumarin), 7.52–7.29 (4H, m, Ar–H), 3.56 (q, $J = 5.9$ Hz, 2H, $–COOCH_2–$), 1.29 (t, $J = 6.6$ Hz, 3H, $–COOCH_2CH_3$).

*Synthesis of 2-oxo-2H-chromene-3-carbohydrazide (**3**)*

2-Oxo-2*H*-chromene-3-carboxylate (5.0 g, 0.023 mol) and hydrazine hydrate 99 % (1.15 g, 0.023 mol) were dissolved in ethanol (100 mL) to give clear solution and refluxed for 10 h. The content was concentrated to half of the volume and allowed to cool. The solid mass thus obtained (reaction was monitored by thin layer chromatography in toluene–acetone solvent system) on cooling was retained by filtering and washed with small amount of ice–cooled ethanol to afford **3** as white solid (Bhat *et al.*, 2008), yield: 88 %, mp 137–140 °C. IR (KBr, cm^{-1}): 3,385, 3,290 (NHs), 1,721 (CO, coumarin), 1,687 (C=O,

amide). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.24 (1H, s, H-4, coumarin), 8.12 (m, 1H, CONHNH$_2$), 7.62–7.39 (4H, m, Ar–H), 4.78 (s, 2H, NH$_2$, D$_2$O exchangeable).

*Synthesis of 3-(5-mercapto-1,3,4-oxadiazol-2-yl)-2H-chromen-2-one (**4**)*

To a solution of the 2-oxo-2*H*-chromene-3-carbohydrazide, **3** (2.5 g, 0.01 mol) in ethanol (50 mL) at 0 °C, carbon disulphide (0.01 mol) and potassium hydroxide (0.01 mol) were added and the reaction mixture was refluxed until the evolution of H_2S gas ceased. Excess solvents were evaporated under reduced pressure and the residue was dissolved in water and then acidified with dilute hydrochloric acid (10 %) to pH 6. The precipitate was filtered off, dried and crystallized from ethanol to give **4** (Patel *et al*., 2010). The completion of reaction was monitored by thin layer chromatography in *n*-hexane–ethyl acetate (8:2) solvent system. Yield 72 %, mp 166–168 °C. IR (KBr, cm^{-1}): 2,591 (SH), 1,726 (CO, coumarin), 1,629, 1,531 (2C=N, oxadiazole), 1,043 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 14.51 (br s, 1H, SH), 8.21 (1H, s, H-4, coumarin), 7.58–7.37 (m, 4H, Ar–H).

General synthetic procedure for 2-(5-2-oxo-2*H*-chromen-3-yl)-1,3,4-oxadiazol-2-ylthio)-*N*-phenylacetamides (**5a–k**)

To a 0.05 mol of 3-(5-mercapto-1,3,4-oxadiazol-2-yl)-2*H*-chromen-2-one (**4**) in 25 mL of acetone, 0.05 mol of 2-chloro-*N*-(substituted)phenyl acetamide (**2a–k**) in acetone and 0.05 mol of K_2CO_3 was added and the reaction mixture was refluxed for 6–12 h, then, after cooling to room temperature, it was slowly poured into 100 mL of ice water and the resulting solid was separated by filtration and washed successively with water. The product was dried under vacuum to obtain **5a–k** (Amir *et al*., 2011). The progress of the reaction was monitored by thin layer chromatography using *n*-hexane–ethyl acetate (8:2) solvent system.

*2-(5-(2-Oxo-2H-chromen-3-yl)-1,3,4-oxadiazol-2-ylthio)-N-phenylacetamide (**5a**)*

Yellow solid, yield 67 %, mp 258–260 °C. IR (KBr, cm^{-1}): 3,308 (NH), 1,723 (CO, coumarin), 1,688 (C=O), 1,630, 1,534 (2C=N, oxadiazole), 1,062 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.42 (s, 1H, –NH), 8.21 (s, 1H, H-4, coumarin), 7.59–7.31 (m, 6H, Ar–H), 4.32 (s, 2H, S-CH$_2$), 7.19 (d, J = 14.6 Hz, 1H), 4.10 (s, 2H); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 170.24 (1C, C-2, oxadiazole), 167.82 (1C, C=O), 164.99 (1C, C-5, oxadiazole), 161.87 (1C, C=O, coumarin), 146.17, 143.13, 141.87, 138.90, 136.45, 133.99, 131.24, 128.78, 125.93, 123.59, 122.21, 119,85, 116.77, 114.89 (14C, Ar–C), 37.13 (1C, S-CH$_2$). Anal. Calcd for $C_{19}H_{13}N_3O_4S$: C 60.15, H 3.45, N 11.08; found: C 59.97, H 3.59, N 10.94).

*N-(4-Chloro-phenyl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**5b**)*

Yellow solid, yield 66 %, mp 265–266 °C. IR (KBr, cm^{-1}): 3,311 (NH), 1,718 (CO, coumarin), 1,681 (C=O), 1,623, 1,524 (2C=N, oxadiazole), 1,075 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.51 (s, 1H, –NH), 8.29 (s, 1H, H-4, coumarin), 7.82 (dd, J = 7.8, 1.3 Hz, 1H), 7.63–7.54 (m, 3H, Ar–H), 7.43–7.28 (m, 4H), 4.23 (s, 2H, S-CH$_2$); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 172.20 (1C, C-2, oxadiazole), 168.22 (1C, C=O), 163.90 (1C, C-5, oxadiazole), 162.39 (1C, C=O, coumarin), 151.24, 148.70, 147.62, 145.91, 142.79, 139.17, 136.98, 135.02, 133.20, 130.38, 127.49, 124.33, 120.68, 118.77 (14C, Ar–C), 31.79 (1C, S-CH$_2$). Anal. Calcd for $C_{19}H_{12}ClN_3O_4S$: C 55.14, H 2.92, N 10.15; found: C 55.29, H 2.78, N 9.96.

*N-(4-Bromo-phenyl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**5c**)*

Yellow solid, yield 71 %, mp 255–257 °C. IR (KBr, cm^{-1}): 3,294 (NH), 1,726 (CO, coumarin), 1,677 (C=O), 1,621, 1,544 (2C=N, oxadiazole), 1,068 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.44 (s, 1H, –NH), 8.25 (s, 1H, H-4, coumarin), 7.83 (dd, J = 7.7, 1.6 Hz, 1H), 7.67–7.55 (m, 3H, Ar–H), 7.46–7.34 (m, 2H), 4.26 (s, 2H, S-CH$_2$), 3.74 (s, 2H, –N–CH$_2$); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 169.30 (1C, C-2, oxadiazole), 166.37 (1C, C=O), 164.02 (1C, C-5, oxadiazole), 163.11 (1C, C=O, coumarin), 148.92, 146.77, 145.90, 144.27, 142.33, 139.88, 137.65, 134.99, 132.14, 129.83, 126.73, 124.32, 123.10, 120.93 (14C, Ar–C), 33.22 (1C, S-CH$_2$). Anal. Calcd for $C_{19}H_{12}BrN_3O_4S$: C 49.80, H 2.64, N 9.17; found: C 49.94, H 2.72, N 9.11.

*N-(4-Fluoro-phenyl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**5d**)*

Yellow solid, yield 68 %, mp 270–272 °C. IR (KBr, cm^{-1}): 3,311 (NH), 1,729 (CO, coumarin), 1,680 (C=O), 1,630, 1,531 (2C=N, oxadiazole), 1,060 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.40 (s, 1H, –NH), 8.29 (s, 1H, H-4, coumarin), 7.79 (dd, J = 7.3, 1.8 Hz, 1H), 7.62–7.28 (m, 5H, Ar–H), 7.16 (t, J = 7.2 Hz, 2H), 4.20 (s, 2H, S-CH$_2$); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 171.16 (1C, C-2, oxadiazole), 166.29 (1C,

C=O), 163.73 (1C, C-5, oxadiazole), 161.14 (1C, C=O, coumarin), 153.28, 150.37, 157.88, 146.04, 143.66, 141.27, 138.73, 135.98, 133.22, 129.74, 126.90, 124.23, 121.93, 118.92 (14C, Ar–C), 36.61 (1C, S-CH_2). Anal. Calcd for $C_{19}H_{12}FN_3O_4S$: C 57.43, H 3.04, N 10.57; found: C 57.31, H 2.97, N 10.43.

*N-(4-Iodo-phenyl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**5e**)*

Light yellow solid, yield 65 %, mp 248–249 °C. IR (KBr, cm^{-1}): 3,308 (NH), 1,731 (CO, coumarin), 1,683 (C=O), 1,628, 1,538 (2C=N, oxadiazole), 1,063 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.53 (s, 1H, –NH), 8.19 (s, 1H, H-4, coumarin), 7.67–7.26 (m, 6H, Ar–H), 7.18 (d, $J = 8.1$ Hz, 2H), 4.27 (s, 2H, S-CH_2); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 168.96 (1C, C-2, oxadiazole), 167.09 (1C, C=O), 163.36 (1C, C-5, oxadiazole), 162.56 (1C, C=O, coumarin), 146.12, 144.87, 143.04, 141.93, 137.99, 136.07, 134.16, 130.82, 128.36, 125.97, 123.23, 120.94, 117.65, 115.79 (14C, Ar–C), 34.09 (1C, S-CH_2). Anal. Calcd for $C_{19}H_{12}IN_3O_4S$: C 45.16, H 2.39, N 8.32; found C 45.03, H 2.49, N 8.21,

*N-(4-Nitro-phenyl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**5f**)*

Yellow solid, yield 61 %, mp 278–280 °C. IR (KBr, cm^{-1}): 3,273 (NH), 1,727 (CO, coumarin), 1,680 (C=O), 1,637, 1,547 (2C=N, oxadiazole), 1,087 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.47 (s, 1H, –NH), 8.22 (s, 1H, H-4, coumarin), 7.74 (d, $J = 7.6$ Hz, 2H), 7.58–7.26 (m, 6H, Ar–H), 4.23 (s, 2H, S-CH_2); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 169.24 (1C, C-2, oxadiazole), 168.11 (1C, C=O), 164.73 (1C, C-5, oxadiazole), 163.01 (1C, C=O, coumarin), 146.23, 144.32, 143.76, 140.93, 137.88, 136.12, 133.56, 131.82, 128.77, 126.21, 125.10, 122.94, 120.32, 119.26 (14C, Ar–C), 36.05 (1C, S-CH_2). Anal. Calcd for $C_{19}H_{12}N_4O_6S$: C 53.77, H 2.85, N 13.20; found: C 53.64, H 2.89, N 13.09.

*N-(4-Cyano-phenyl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**5g**)*

Yellow solid, yield 67 %, mp 243–245 °C. IR (KBr, cm^{-1}): 3,279 (NH), 1,717 (CO, coumarin), 1675 (C=O), 1621, 1531 (2C=N, oxadiazole), 1067 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.39 (s, 1H, –NH), 8.30 (s, 1H, H-4, coumarin), 7.78 (d, $J = 7.3$ Hz, 2H), 7.66 (d, $J = 7.4$ Hz, 2H), 7.53–7.27 (m, 4H, Ar–H), 4.26 (s, 2H, S-CH_2); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 170.79 (1C, C-2, oxadiazole), 166.87 (1C, C=O), 165.21 (1C, C-5, oxadiazole), 162.29 (1C, C=O, coumarin), 148.19, 147.81, 145.17, 143.55, 141.78, 137.68, 136.02, 133.42, 130.28, 126.77, 124.35, 121.79, 117.69 (13C, Ar–C), 104.98 (1C, $\underline{C}\equiv N$), 96.91 (1C, –$\underline{C}$–$C\equiv N$), 33.90 (1C, S-CH_2). Anal. Calcd for $C_{20}H_{12}N_4O_4S$: C 59.40, H 2.99, N 13.85; found: C 59.53, H 3.07, N 13.71.

*2-[5-(2-Oxo-2H–2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-N-p-tolyl-acetamide (**5h**)*

Light yellow solid, yield 74 %, mp 254–255 °C. IR (KBr, cm^{-1}): 3,321 (NH), 1,727 (CO, coumarin), 1,675 (C=O), 1,636, 1,541 (2C=N, oxadiazole), 1,070 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.55 (s, 1H, –NH), 8.35 (s, 1H, H-4, coumarin), 7.70–7.32 (m, 8H, Ar–H), 4.20 (s, 2H, S-CH_2), 1.33 (s, 3H, CH_3); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 172.56 (1C, C-2, oxadiazole), 168.27 (1C, C=O), 163.88 (1C, C-5, oxadiazole), 161.99 (1C, C=O, coumarin), 150.92, 148.70, 147.32, 144.58, 142.31, 138.70, 134.88, 131.28, 129.83, 125.69, 124.10, 121.28, 118.93, 115.77 (14C, Ar–C), 33.98 (1C, S-CH_2), 19.87 (1C, CH_3). Anal. Calcd for $C_{20}H_{15}N_3O_4S$: C 61.06, H 3.84, N 10.68; found: C 61.21, H 3.70, N 10.53.

*N-(4-Methoxy-phenyl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**5i**)*

Yellow solid, mp 252–255 °C, yield 69 %. IR (KBr, cm^{-1}): 3,298 (NH), 1,727 (CO, coumarin), 1,683 (C=O), 1,633, 1,530 (2C=N, oxadiazole), 1,067 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.57 (s, 1H, –NH), 8.32 (s, 1H, H-4, coumarin), 7.77 (d, $J = 7.1$ Hz, 2H), 7.57–7.30 (m, 4H, Ar–H), 6.93 (d, $J = 7.4$ Hz, 2H), 4.23 (s, 2H, S-CH_2), 3.86 (s, 3H, –OCH_3); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 169.66 (1C, C-2, oxadiazole), 168.13 (1C, C=O), 163.80 (1C, C-5, oxadiazole), 161.92 (1C, C=O, coumarin), 147.67, 146.13, 143.87, 142.13, 140.72, 136.89, 135.60, 132.17, 129.99, 127.65, 126.27, 124.70, 122.27, 119.57 (14C, Ar–C), 55.76 (1C, OCH_3), 35.80 (1C, S-CH_2). Anal. Calcd for $C_{20}H_{15}N_3O_5S$: C 58.67, H 3.69, N 10.26; found: C 58.78, H 3.61, N 10.37.

*N-(4-Ethoxy-phenyl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**5j**)*

Yellow solid, yield 63 %, mp 260–262 °C. IR (KBr, cm^{-1}): 3,289 (NH), 1,729 (CO, coumarin), 1,680 (C=O), 1,632, 1,539 (2C=N, oxadiazole), 1,057 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.52 (s, 1H, –NH), 8.26 (s, 1H, H-4, coumarin), 7.80 (d, $J = 7.3$ Hz, 2H), 7.62–7.29 (m, 4H, Ar–H), 6.88 (d, $J = 7.8$ Hz, 2H), 4.17 (s, 2H, S-CH_2), 3.90 (q, $J = 6.7$ Hz, 2H, –O–CH_2–), 1.23 (t, $J = 5.7$ Hz, 3H, CH_3); ^{13}C NMR

(100 MHz, DMSO-d_6): δ ppm, 170.24 (1C, C-2, oxadiazole), 167.80 (1C, C=O), 164.16 (1C, C-5, oxadiazole), 162.54 (1C, C=O, coumarin), 149.87, 147.69, 146.27, 143.88, 140.24, 137.66, 136.71, 133.23, 132.98, 128.99, 127.53, 124.33, 121.93, 118.77 (14C, Ar–C), 65.61 (1C, OCH_2CH_3), 35.32 (1C, S-CH_2), 22.31 (1C, OCH_2CH_3). Anal. Calcd for $C_{21}H_{17}N_3O_5S$: C 59.57, H 4.05, N 9.92; found: C 59.71, H 3.91, N 10.04.

*N-(4-Acetylamino-phenyl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**5k**)*

Yellow solid, yield 65 %, mp 239–241 °C. IR (KBr, cm^{-1}): 3,299 (NH), 1,727 (CO, coumarin), 1,685 (C=O), 1,623, 1,530 (2C=N, oxadiazole), 1,072 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.59 (s, 1H, –NH), 8.42 (s, 1H, –NH), 8.22 (s, 1H, H-4, coumarin), 7.73 (d, $J = 7.6$ Hz, 2H), 7.62–7.28 (m, 6H, Ar–H), 4.26 (s, 2H, S-CH_2), 1.86 (s, 3H, –CH_3); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 169.82 (1C, C-2, oxadiazole), 167.54, 166.44 (2C, C=O), 163.90 (1C, C-5, oxadiazole), 161.81 (1C, C=O, coumarin), 146.50, 144.32, 143.91, 141.27, 137.89, 136.15, 133.49, 131.92, 129.74, 126.55, 124.32, 123.29, 121.75, 118.73 (14C, Ar–C), 34.33 (1C, S-CH_2), 19.65 (1C, CH_3). Anal. Calcd for $C_{21}H_{16}N_4O_5S$: C 57.79, H 3.70, N 12.84; found: C 57.89, H 3.61, N 12.69.

General synthetic procedure for *N*-(benzo[d]thiazol-2-yl)-2-(5-(2-oxo-2*H*-chromen-3-yl)-1,3,4-oxadiazol-2-ylthio)acetamides (**6a–k**)

To a 0.05 mol of 3-(5-mercapto-1,3,4-oxadiazol-2-yl)-2*H*-chromen-2-one (**4**) in 25 mL of acetone, 0.05 mol of *N*-(benzo[d]thiazol-2-yl)-2-chloroacetamides (**4a–k**) in acetone and 0.05 mol of K_2CO_3 was added and the reaction mixture was refluxed for 8–15 h, then, after cooling to room temperature, it was slowly poured into 100 mL of ice water and the resulting solid was separated by filtration and washed successively with water. The product was dried under vacuum to obtain **6a–k** (Akhtar *et al*., 2008). The progress of the reaction was monitored by thin layer chromatography using *n*-hexane–ethyl acetate (8:2) solvent system.

*N-(Benzo[d]thiazol-2-yl)-2-(5-(2-oxo-2H-chromen-3-yl)-1,3,4-oxadiazol-2-ylthio)acetamide (**6a**)*

Yellow solid, yield 68 %, mp 254–256 °C. IR (KBr, cm^{-1}): 3,321 (NH), 1,723 (CO, coumarin), 1,682 (C=O), 1,654 (C=N, benzothiazole), 1,626, 1,533 (2C=N, oxadiazole), 1,081 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.53 (s, 1H, –NH), 8.19 (s, 1H, H-4, coumarin), 7.66–7.21 (m, 8H, Ar–H), 4.24 (s, 2H, S-CH_2); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 170.24 (1C, C-2, oxadiazole), 167.82 (1C, C=O), 164.99 (1C, C-5, oxadiazole), 161.87 (1C, C=O, coumarin), 155.98 (1C, C-2, benzothiazole), 148.92, 146.34, 143.67, 140.96, 138.65, 137.03, 134.99, 131.26, 128.60, 126.63, 125.55, 123.20, 120.12, 119.99 (14C, Ar–C), 36.16 (1C, S-CH_2). Anal. Calcd for $C_{20}H_{12}N_4O_4S_2$: C 55.04, H 2.77, N 12.84; found: C 55.22, H 2.59, N 12.96.

*N-(6-Chloro-benzothiazol-2-yl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**6b**)*

Yellow solid, yield 74 %, mp 261–263 °C. IR (KBr, cm^{-1}): 3,285 (NH), 1,728 (CO, coumarin), 1,673 (C=O), 1,657 (C=N, benzothiazole), 1,635, 1,530 (2C=N, oxadiazole), 1,073 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.59 (s, 1H, –NH), 8.22 (s, 1H, H-4, coumarin), 7.82 (d, $J = 1.7$ Hz, 1H), 7.53 (d, $J = 7.3$ Hz, 1H), 7.41 (dd, $J = 7.5$, 1.6 Hz, 1H), 7.34–7.17 (m, 4H, Ar–H), 4.31 (s, 2H, S-CH_2); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 171.83 (1C, C-2, oxadiazole), 169.18 (1C, C=O), 165.73 (1C, C-5, oxadiazole), 162.19 (1C, C=O, coumarin), 153.61 (1C, C-2, benzothiazole), 148.79, 146.21, 145.03, 143.29, 140.82, 137.65, 135.34, 131.95, 128.74, 127.52, 124.89, 121.86, 119.26, 117.60 (14C, Ar–C), 36.22 (1C, S-CH_2). Anal. Calcd for $C_{20}H_{11}ClN_4O_4S_2$: C 51.01, H 2.35, N 11.90; found: C 50.87, H 2.49, N 11.76.

*N-(6-Bromo-benzothiazol-2-yl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**6c**)*

Yellow solid, yield 71 %, mp 256–258 °C. IR (KBr, cm^{-1}): 3,313 (NH), 1,719 (CO, coumarin), 1,675 (C=O), 1,661 (C=N, benzothiazole), 1,631, 1,524 (2C=N, oxadiazole), 1,065 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.51 (s, 1H, –NH), 8.26 (s, 1H, H-4, coumarin), 7.73 (d, $J = 1.6$ Hz, 1H), 7.56–7.19 (m, 6H, Ar–H), 4.32 (s, 2H, S-CH_2); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 169.82 (1C, C-2, oxadiazole), 166.69 (1C, C=O), 165.03 (1C, C-5, oxadiazole), 163.10 (1C, C=O, coumarin), 153.22 (1C, C-2, benzothiazole), 146.60, 144.32, 143.20, 140.92, 137.89, 136.02, 133.85, 131.92, 128.76, 126.45, 124.39, 121.56, 117.69, 116.29 (14C, Ar–C), 37.11 (1C, S-CH_2). Anal. Calcd for $C_{20}H_{11}BrN_4O_4S_2$: C 46.61, H 2.15, N 10.87; found: C 46.47, H 2.22, N 10.77.

*N-(6-Fluoro-benzothiazol-2-yl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**6d**)*

Yellow solid, yield 62 %, mp 274–275 °C. IR (KBr, cm^{-1}): 3,286 (NH), 1,727 (CO, coumarin), 1,680 (C=O), 1,659 (C=N, benzothiazole), 1,629, 1,527 (2C=N, oxadiazole), 1,073 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz,

DMSO-d_6): δ ppm, 8.48 (s, 1H, –NH), 8.25 (s, 1H, H-4, coumarin), 7.79 (dd, $J = 8.2$, 1.6 Hz, 1H), 7.61 (dd, $J = 2.0$, 8.3 Hz, 1H), 7.42 (dd, $J = 5.2$, 8.1 Hz, 1H), 7.30–7.16 (m, 4H, Ar–H), 4.30 (s, 2H, S-CH_2); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 170.52 (1C, C-2, oxadiazole), 168.09 (1C, C=O), 166.25 (1C, C-5, oxadiazole), 162.81 (1C, C=O, coumarin), 152.51 (1C, C-2, benzothiazole), 145.41, 144.22, 141.93, 139.24, 136.58, 134.23, 133.28, 130.92, 128.68, 126.45, 125.20, 123.20, 120.84, 119.37 (14C, Ar–C), 35.62 (1C, S-CH_2). Anal. Calcd for $C_{20}H_{11}FN_4O_4S_2$: C 52.86, H 2.44, N 12.33 %; found: C 52.99, H 2.53, N 12.21.

*N-(6-Iodo-benzothiazol-2-yl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**6e**)*

Yellow solid, yield 69 %, mp 250–253 °C. IR (KBr, cm^{-1}): 3,394 (NH), 1,731 (CO, coumarin), 1,676 (C=O), 1,652 (C=N, benzothiazole), 1,621, 1,539 (2C=N, oxadiazole), 1,078 (C–O–C, oxadiazole). ^{1}H. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.52 (s, 1H, –NH), 8.29 (s, 1H, H-4, coumarin), 7.86 (d, $J = 2.3$ Hz, 1H), 7.71 (d, $J = 7.7$ Hz, 1H), 7.51 (dd, $J = 8.6$, 1.6 Hz, 1H), 7.38–7.23 (m, 4H, Ar–H), 4.33 (s, 2H, S-CH_2); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 169.37 (1C, C-2, oxadiazole), 166.29 (1C, C=O), 163.92 (1C, C-5, oxadiazole), 161.83 (1C, C=O, coumarin), 154.70 (1C, C-2, benzothiazole), 146.24, 145.20, 142.79, 140.38, 138.75, 135.90, 132.76, 131.28, 129.05, 126.77, 125.28, 122.75, 120.34, 117.85 (14C, Ar–C), 33.84 (1C, S-CH_2). Anal. Calcd for $C_{20}H_{11}IN_4O_4S_2$: C 42.72, H 1.97, N 9.96; found: C 42.61, H 2.11, N 9.83.

*N-(6-Nitro-benzothiazol-2-yl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**6f**)*

Yellow solid, yield 68 %, mp 291–293 °C. IR (KBr, cm^{-1}): 3,308 (NH), 1,726 (CO, coumarin), 1,669 (C=O), 1,655 (C=N, benzothiazole), 1,620, 1,525 (2C=N, oxadiazole), 1,072 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.53 (s, 1H, –NH), 8.20 (s, 1H, H-4, coumarin), 7.91 (d, $J = 1.6$ Hz, 1H), 7.74 (dd, $J = 7.1$, 2.1 Hz, 1H), 7.59 (d, $J = 7.4$ Hz, 1H), 7.43 (dd, $J = 7.5$, 1.4 Hz, 2H), 7.34–7.26 (m, 2H, Ar–H), 4.18 (s, 2H, S-CH_2); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 169.24 (1C, C-2, oxadiazole), 167.59 (1C, C=O), 165.14 (1C, C-5, oxadiazole), 162.53 (1C, C=O, coumarin), 151.98 (1C, C-2, benzothiazole), 147.45, 146.24, 144.39, 141.89, 137.69, 135.79, 134.27, 131.93, 128.73, 125.96, 123.28, 121.73, 120.95, 117.63 (14C, Ar–C), 36.70 (1C, S-CH_2). Anal. Calcd for $C_{20}H_{11}N_5O_6S_2$: C 49.89, H 2.30, N 14.55; found: C 50.05, H 2.13, N 14.59.

*N-(6-Cyano-benzothiazol-2-yl)-2-[5-(2-oxo-2H–2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**6g**)*

Yellow solid, mp 260–262 °C, yield 63 %. IR (KBr, cm^{-1}): 3,318 (NH), 1,727 (CO, coumarin), 1,688 (C=O), 1,649 (C=N, benzothiazole), 1,635, 1,541 (2C=N, oxadiazole), 1,080 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.55 (s, 1H, –NH), 8.21 (s, 1H, H-4, coumarin), 7.84 (d, $J = 1.8$ Hz, 1H), 7.68 (d, $J = 7.5$ Hz, 1H), 7.45 (dd, $J = 8.7$, 1.6 Hz, 1H), 7.37–7.24 (m, 4H, Ar–H), 4.33 (s, 2H, S-CH_2); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 172.33 (1C, C-2, oxadiazole), 168.23 (1C, C=O), 165.47 (1C, C-5, oxadiazole), 162.92 (1C, C=O, coumarin), 154.19 (1C, C-2, benzothiazole), 146.23, 144.34, 141.29, 140.27, 138.76, 137.20, 134.89, 132.15, 130.26, 126.79, 125.47, 122.47, 117.89 (13C, Ar–C), 106.16 (1C, C $\equiv$ N), 97.57 (1C, –C–C $\equiv$ N), 34.13 (1C, S-CH_2). Anal. Calcd for $C_{21}H_{11}N_5O_4S_2$: C 54.66, H 2.40, N 15.18; found: C 54.78, H 2.31, N 15.03.

*N-(6-Methyl-benzothiazol-2-yl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**6h**)*

Yellow solid, yield 73 %, mp 246–247 °C. IR (KBr, cm^{-1}): 3,284 (NH), 1,729 (CO, coumarin), 1,675 (C=O), 1,662 (C=N, benzothiazole), 1,630, 1,522 (2C=N, oxadiazole), 1,063 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.53 (s, 1H, –NH), 8.28 (s, 1H, H-4, coumarin), 7.77 (d, $J = 7.8$ Hz, 1H), 7.64–7.23 (m, 6H, Ar–H), 4.36 (s, 2H, S-CH_2), 1.39 (s, 3H, CH_3); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 169.56 (1C, C-2, oxadiazole), 167.27 (1C, C=O), 165.28 (1C, C-5, oxadiazole), 161.90 (1C, C=O, coumarin), 153.21 (1C, C-2, benzothiazole), 145.87, 143.28, 140.92, 138.66, 136.54, 135.40, 132.98, 130.28, 127.86, 124.94, 123.09, 120.83, 118.73, 116.54 (14C, Ar–C), 36.02 (1C, S-CH_2), 19.80 (1C, CH_3). Anal. Calcd for $C_{21}H_{14}N_4O_4S_2$: C 55.99, H 3.13, N 12.44; found: C 56.14, H 2.96, N 12.31.

*N-(6-Methoxy-benzothiazol-2-yl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**6i**)*

Yellow solid, yield 67 %, mp 268–270 °C. IR (KBr, cm^{-1}): 3,297 (NH), 1,723 (CO, coumarin), 1,688 (C=O), 1,666 (C=N, benzothiazole), 1,624, 1,539 (2C=N, oxadiazole), 1,060 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.62 (s, 1H, –NH), 8.29 (s, 1H, H-4, coumarin), 7.83–7.72 (m, 2H), 7.48 (dd, $J = 8.5$, 1.8 Hz, 1H), 7.38–7.23 (m, 4H, Ar–H), 4.27 (s, 2H, S-CH_2), 3.78 (s, 3H, OCH_3); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 170.09 (1C, C-2, oxadiazole), 168.92 (1C, C=O), 165.15 (1C, C-5,

oxadiazole), 162.92 (1C, C=O, coumarin), 150.92 (1C, C-2, benzothiazole), 146.57, 143.29, 142.39, 140.92, 138.76, 135.99, 134.20, 130.94, 127.89, 126.54, 125.32, 123.95, 120.63, 118.73 (14C, Ar–C), 56.38 (1C, OCH_3), 36.19 (1C, S-CH_2). Anal. Calcd for $C_{21}H_{14}N_4O_5S_2$: C 54.07, H 3.02, N 12.01; found: C 54.22, H 3.19, N 11.88.

*N-(6-Ethoxy-benzothiazol-2-yl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**6j**)*

Yellow solid, yield 71 %, mp 275–277 °C. IR (KBr, cm^{-1}): 3,316 (NH), 1,732 (CO, coumarin), 1,673 (C=O), 1,658 (C=N, benzothiazole), 1,620, 1,541 (2C=N, oxadiazole), 1,077 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.50 (s, 1H, –NH), 8.25 (s, 1H, H-4, coumarin), 7.79–7.67 (m, 2H), 7.57 (dd, $J = 8.4$, 1.9 Hz, 1H), 7.38–7.26 (m, 4H, Ar–H), 4.35 (s, 2H, S-CH_2), 4.14 (q, $J = 6.6$ Hz, 2H, OCH_2CH_3), 2.21 (t, $J = 6.7$ Hz, 3H, CH_2CH_3); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 169.87 (1C, C-2, oxadiazole), 167.03 (1C, C=O), 164.73 (1C, C-5, oxadiazole), 161.29 (1C, C=O, coumarin), 155.12 (1C, C-2, benzothiazole), 145.46, 144.20, 140.94, 137.89, 135.69, 134.32, 133.28, 130.92, 128.97, 127.64, 125.69, 123.41, 122.94, 119.73 (14C, Ar–C), 65.19 (1C, OCH_2CH_3), 35.03 (1C, S-CH_2), 22.19 (1C, OCH_2CH_3). Anal. Calcd for $C_{22}H_{16}N_4O_5S_2$: C 54.99, H 3.36, N 11.66; found: C 55.16, H 3.30, N 11.52.

*N-(6-Acetylamino-benzothiazol-2-yl)-2-[5-(2-oxo-2H-chromen-3-yl)-[1,3,4]oxadiazol-2-ylsulfanyl]-acetamide (**6k**)*

Yellow solid, yield 66 %, mp 257–259 °C. IR (KBr, cm^{-1}): 3,289 (NH), 1,728 (CO, coumarin), 1,682 (C=O), 1,655 (C=N, benzothiazole), 1,624, 1,530 (2C=N, oxadiazole), 1,081 (C–O–C, oxadiazole). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm, 8.59 (s, 1H, –NH), 8.42 (s, 1H, –NH), 8.28 (s, 1H, H-4, coumarin), 7.79 (d, $J = 1.6$ Hz, 2H), 7.64 (dd, $J = 7.8$, 1.9 Hz, 2H), 7.59 (dd, $J = 8.6$, 1.7 Hz, 1H), 7.40–7.31 (m, 2H, Ar–H), 4.33 (s, 2H, S-CH_2), 2.19 (s, 3H, CH_3); ^{13}C NMR (100 MHz, DMSO-d_6): δ ppm, 170.76 (1C, C-2, oxadiazole), 168.32, 167.84 (2C, CO), 164.90 (1C, C-5, oxadiazole), 161.82 (1C, C=O, coumarin), 154.21 (1C, C-2, benzothiazole), 146.78, 145.70, 143.25, 141.28, 140.10, 138.76, 135.48, 132.97, 130.92, 127.99, 126.25, 123.76, 120.93, 116.54 (14C, Ar–C), 36.34 (1C, S-CH_2), 19.35 (1C, CH_3). Anal. Calcd for $C_{22}H_{15}N_5O_5S_2$: C 53.54, H 3.06, N 14.19; found: C 53.40, H 2.94, N 14.04.

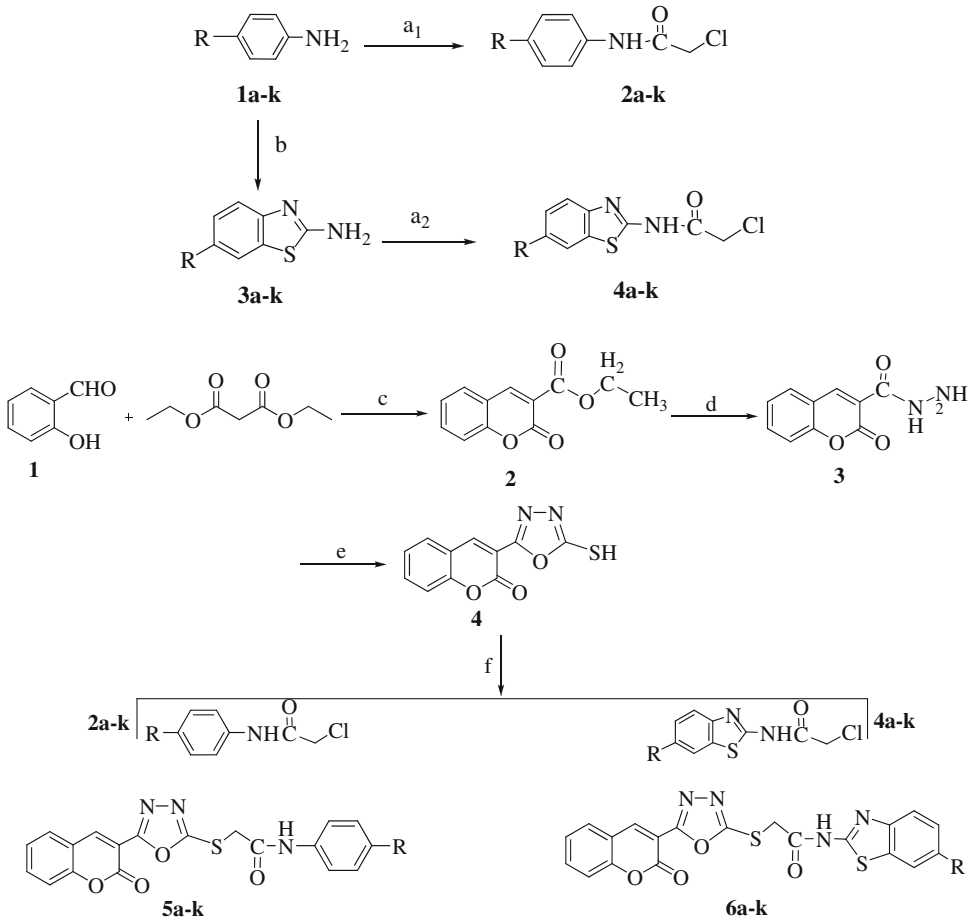
Methods for pharmacological activity evaluations

The synthesized derivatives (**5a–k**, **6a–k**) were examined for antimicrobial activity against several bacteria (*Staphylococcus aureus* MTCC 96, *Bacillus cereus* MTCC 430, *Escherichia coli* MTCC 739, *Pseudomonas aeruginosa* MTCC 741, *Klebsiella pneumoniae* MTCC 109, *Salmonella typhi* MTCC 733, *Proteus vulgaris* MTCC 1771, *Shigella flexneri* MTCC 1457), fungi (*Aspergillus niger* MTCC 282, *Aspergillus fumigatus* MTCC 343, *Aspergillus clavatus* MTCC 1323, *Candida albicans* MTCC 183) using agar streak dilution method as well as against *M. tuberculosis* H37Rv strain using BACTEC MGIT and Lowenstein–Jensen MIC method as reported earlier (Hawkey and Lewis, 1994; Anargyros *et al*., 1990; Isenberg, 1992; Patel *et al*., 2011c, d). Ciprofloxacin and ketoconazole were used as control drugs for antibacterial and antifungal activities, respectively, whereas isoniazid, rifampicin, ethambutol and pyrazinamide are used as control drugs for antituberculosis activity.

Results and discussion

Scheme 1 outlines the synthetic pathway followed by the synthesis of the title compounds **5a–k** and **6a–k**. The solvents and reagents were used as received or were dried prior to use as needed. Salicylaldehyde and diethylmalonate were reacted in the presence of piperidine in ethanol to form ethyl-2-oxo-2*H*-chromene-3-carboxylate (**2**) which on treatment with 99 % hydrazine hydrate yielded 2-oxo-2*H*-chromonene-3-carbohydrazide (**3**) in good yield. The resulted carbohydrazide moiety was cyclized using sodium hydroxide in the presence of carbon disulphide to furnish the corresponding oxadiazole nucleus, 3-(5-sulfanyl-1,3,4-oxadiazol-2-yl)-2*H*-chromen-2–one (**4**). 2-Chloro-*N*-phenyl acetamides were synthesized according to the reported literature (Baraldi *et al*., 2007). Chloroacetyl chloride was reacted with *para*-(substituted) phenyl amines to give the corresponding 2-chloro-*N*-(substituted)phenyl acetamides (**2a–k**) and the correct synthesis was confirmed using FT–IR and ^{1}H NMR spectroscopy. The 2-amino-6-substituted benzothiazoles (**3a–k**) were synthesized by reacting aryl amines with potassium thiocyanates in satisfactory yields by a known preparation method (Wang *et al*., 2006; Rana *et al*., 2008) and were converted to the respective *N*-(benzo[d]thiazol-2-yl)-2-chloroacetamides (**4a–k**) using chloroacetyl chloride in benzene solvent as described in the literature (Turan-Zitouni *et al*., 2003; Amin *et al*., 2008).

Above-mentioned acetamide derivatives (**2a–k** and **4a–k**) were then condensed to the 3-(5-mercapto-1,3,4-oxadiazol-2-yl)-2*H*-chromen-2–one (**4**) in acetone solvent at reflux temperature furnished **5a–k** and **6a–k**. All the IR, ^{1}H NMR, ^{13}C NMR spectral data of compounds **5a–k** and **6a–k** were in accordance with assumed structures. The purity of the synthesized compounds was monitored by TLC and ascertained by elemental analysis.



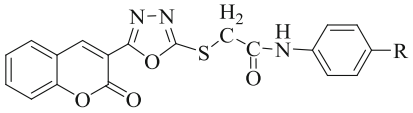
Scheme 1 Synthetic procedure for **5a–k** and **6a–k.** Reagents and conditions: (a_1) $ClCH_2COCl$, anhyd. K_2CO_3, acetone; (a_2) $ClCH_2COCl$, anhyd. K_2CO_3, benzene; (*b*) KSCN, Br_2, AcOH, R.T.; (*c*) piperidine, reflux, EtOH; (*d*) $NHNH_2{\cdot}H_2O$, reflux, EtOH; (*e*) CS_2/KOH, reflux, EtOH; (*f*) Acetone, reflux

Biological activity

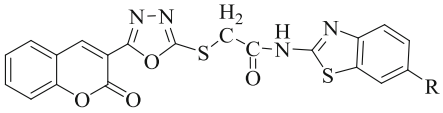
Investigation on antibacterial screening data (Table 1) showed that some of the compounds showed excellent activity against all the mentioned bacteria. From the bioassay it can be stated that the final analogues with the substitutions of *N*-benzothiazolyl-2-chloroacetamides were, in most cases, active against all the pathogenic strains studied than the analogues bearing 2-chloro-*N*-phenyl acetamides. In addition, final analogues exhibited good antibacterial activity against Gram-positive bacteria when compared to that against Gram-negative ones.

Final analogue **5c** with 2-chloro-*N*-phenyl acetamide constituent bearing electron-donating halogen atom in the form of bromine, showed higher effectiveness against Gram-negative strain *E. coli* at 12.5 μg/mL of MIC, while another analogue (**5g**) from the same series bearing electron-withdrawing cyano substituent indicated excellent inhibitory efficacy of Gram-positive bacteria *S. aureus* at 6.25 μg/mL of MIC along with similar efficacy of analogue **5i** bearing electron-withdrawing methoxy functional group against the same bacteria. The latter analogue (**5i**) was the only and most potent analogue against Gram-negative strain *S. typhi* at 25 μg/mL of MIC, whereas similar to previously mentioned analogue **5g**, analogue **6g** with similar cyano functional group incorporated to the benzothiazole ring system showed good level of inhibitory potential against Gram-negative *K. pneumoniae* at 25 μg/mL of MIC. Higher potency (MIC 3.12 μg/mL) has been observed with the analogue **6c** within the class of compounds those involving 2-chloro-*N*-benzothiazole acetamide linkage to the 1,3,4-oxadiazole core towards Gram-positive strain *B. cereus*. The said analogue was the only one showing highest antibacterial activity with lowest MIC value in the present bioassay. Final derivative **5j** with ethoxy functional group indicated significant activity against Gram-negative *P. aeruginosa* at lowest 6.25 μg/mL of MIC and it will be appropriate to mention here that this was the lowest MIC level observed against the mentioned Gram-negative strain, i.e. higher activity level. Furthermore, an analogue **6k** with acetamido functionality showed similar inhibitory efficacy (MIC 6.25 μg/mL) against the same bacteria *P. aeruginosa*. Insertion of electron-withdrawing alkoxy functional group in the form of methoxy functionality to the benzothiazole ting system (**6i**) attached to the nucleus

Table 1 In vitro antibacterial activity of newly synthesized compounds



5a-k



6a-k

Entry	R	Log*P*	MIC (μg/mL)							
			S.a	*B.c*	*E.c*	*P.a*	*K.p*	*S.t*	*P.v*	*S.f*
5a	H	3.08	>100	100	100	>100	>100	>100	>100	>100
5b	Cl	3.64	50	25	25	100	100	100	100	50
5c	Br	3.91	50	12.5	12.5	50	100	100	100	100
5d	F	3.24	12.5	50	50	100	100	50	100	50
5e	I	4.43	100	100	100	100	100	>100	50	100
5f	NO_2	3.11	100	100	50	>100	100	100	25	100
5g	CN	3.11	6.25	100	50	100	50	100	100	>100
5h	CH_3	3.56	100	50	>100	>100	>100	100	>100	>100
5i	OCH_3	2.95	6.25	100	50	25	100	25	100	100
5j	OC_2H_5	3.29	25	100	50	6.25	100	50	100	100
5k	$NHCOCH_3$	1.99	50	100	>100	50	>100	>100	>100	>100
6a	H	4.27	100	100	>100	50	>100	100	100	100
6b	Cl	4.83	50	6.25	50	100	100	50	50	25
6c	Br	5.10	25	3.12	50	100	100	100	50	50
6d	F	4.43	12.5	12.5	50	50	100	50	100	25
6e	I	5.63	100	100	50	>100	100	100	>100	100
6f	NO_2	4.23	50	100	100	100	50	100	12.5	100
6g	CN	4.31	25	100	50	>100	25	100	100	>100
6h	CH_3	4.76	100	100	>100	100	100	100	100	>100
6i	OCH_3	4.15	25	50	100	50	100	25	25	50
6j	OC_2H_5	4.49	12.5	25	50	6.25	100	12.5	100	100
6k	$NHCOCH_3$	3.18	50	100	100	12.5	100	100	>100	100
Cip.			1.56	0.39	1.56	0.78	0.78	0.39	0.39	1.56
DMSO			–	–	–	–	–	–	–	–

Log*P* was calculated using the ChemDraw Ultra, version 11.0

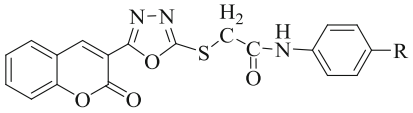
Cip. ciprofloxacin, *S.a Staphylococcus aureus*, *B.c Bacillus cereus*, *E.c Escherichia coli*, *P.a Pseudomonas aeruginosa*, *K.p Klebsiella pneumoniae*, *S.t Salmonella typhi*, *P.v Proteus vulgaris*, *S.f Shigella flexneri*

was found essential to contribute grater activity against Gram-negative *S. typhi* at 25 μg/mL of MIC. Similar inhibitory concentration (25 μg/mL) was observed for analogue **6b** bearing electron-donating halogen (Cl) atom within the series involving benzothiazole ring system against Gram-negative strain *S. flexneri*. Nitro group (strong electron withdrawing) bearing analogue (**6f**) within the benzothiazole ring series showed remarkable activity against Gram-negative strain at 12.5 μg/mL of MIC. Many of the remaining analogues were succeeded to indicate good-to-moderate activity level of 6.25–25 μg/mL against Gram-positive strains as well as 12.5–50 μg/mL against Gram-negative strains, while some analogues were found to indicate poor activity at concentration level 50–100 μg/mL.

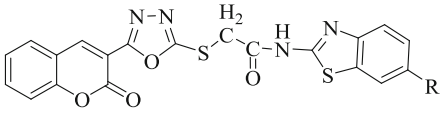
In vitro antifungal screening results for the final analogues **5a**–**6k** are furnished in Table 2 and from the results it can be stated that the analogues displayed comparatively good in vitro antifungal potency (12.5–25 μg/mL) when compared to that of against Gram-positive strains (6.25, 12.5 and 25 μg/mL) as mentioned in the previous table. In addition, it will suffice to mention here that opposite to the antibacterial results, in case of bioassay against fungal strains, halogen substituents (Cl, Br, F) were found more effective as compared to the analogues bearing alkoxy (OCH_3, OC_2H_5) functional groups.

Final analogues bearing benzothiazole ring system involving substitution of electron-donating chloro (**6b**) and fluoro (**6d**) functional groups were found to demonstrate

Table 2 In vitro antifungal activity of newly synthesized compounds



5a-k



6a-k

Entry	R	LogP	MIC (μg/mL)			
			A.n	*A.f*	*A.c*	*C.a*
5a	H	3.08	>100	>100	>100	100
5b	Cl	3.64	25	25	100	50
5c	Br	3.91	50	100	100	50
5d	F	3.24	50	50	100	25
5e	I	4.43	100	100	100	100
5f	NO_2	3.11	100	50	100	12.5
5g	CN	3.11	>100	>100	>100	50
5h	CH_3	3.56	100	>100	100	>100
5i	OCH_3	2.95	100	100	50	100
5j	OC_2H_5	3.29	100	100	100	50
5k	$NHCOCH_3$	1.99	>100	>100	50	100
6a	H	4.27	>100	>100	>100	>100
6b	Cl	4.83	12.5	25	100	100
6c	Br	5.10	25	100	100	50
6d	F	4.43	12.5	25	50	50
6e	I	5.63	100	100	100	100
6f	NO_2	4.23	50	100	100	50
6g	CN	4.31	100	>100	100	50
6h	CH_3	4.76	>100	>100	100	100
6i	OCH_3	4.15	50	100	25	50
6j	OC_2H_5	4.49	100	100	50	100
6k	$NHCOCH_3$	3.18	100	>100	12.5	>100
Kit.			1.56	0.78	0.78	1.56
DMSO			–	–	–	–

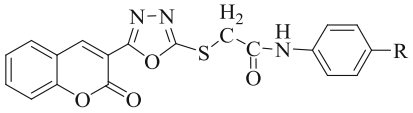
LogP was calculated using the ChemDraw Ultra, version 11.0

Kit ketoconazole, *A.n Aspergillus niger*, *A.f* Aspergillus fumigates, *A.c Aspergillus clavatus*, *C.a Candida albicans*

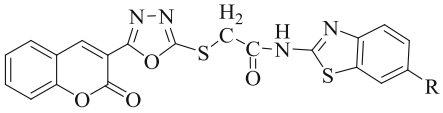
promising activity against two fungal species as *A. niger* and *A. fumigatus* at 12.5 μg/mL and 25 μg/mL of MICs, respectively, along with similar efficacy (25 μg/mL) of analogue **5b** from phenyl acetamide series bearing chlorine substituent against *A. fumigatus*. Analogue **6k** from benzothiazolyl series bearing highly lipophilic acetamido functional group linkage was appeared with noticeable inhibitory potential against *A. clavatus* fungi at 12.5 μg/mL of MIC, while similar inhibitory concentration (12.5 μg/mL) was observed for analogue **5f** bearing strong electron-withdrawing nitro functional group within the series involving substitution of *N*-phenyl acetamide against *C. albicans* fungi. Many of the remaining derivatives were found to indicate moderate inhibition of the mentioned panel of fungal species at 25–50 μg/mL of MIC, whereas some derivatives were shown poor inhibition at 100 μg/mL.

In vitro TB activities of compounds **5a**–**6k** were assessed against *M. tuberculosis* H37Rv (Table 3). Preliminarily, the analogues presented here are examined for their in vitro antituberculosis activity using BACTEC MGIT method at concentration level of 6.25 μg/mL and it is seen from the results that none of the analogues showed activity at such level. Hence, all the analogues were further tested for the same activity using Lowenstein–Jensen MIC method. From the secondary bioassay results is can be stated that both the class of analogues either involving phenyl or benzothiazolyl-acetamide linkages shown equal degree of lowest MICs in the range 12.5–62.5 μg/mL. Two of the final analogues halo substituents as **5e** bearing iodo substituent to the phenyl acetamide moiety and **6b** bearing bromo substituent to the benzothiazolyl acetamide moiety

Table 3 In vitro antituberculosis activity of newly synthesized compounds



5a-k



6a-k

Entry	R	BACTEC MGIT method[a]		L–J MIC method[a]	
		MIC (μg/mL)	% Inhibition	MIC (μg/mL)	% Inhibition
5a	H	>6.25	–	500	95
5b	Cl	>6.25	–	100	96
5c	Br	>6.25	–	62.5	97
5d	F	>6.25	–	25	99
5e	I	>6.25	–	12.5	99
5f	NO_2	>6.25	–	500	94
5g	CN	>6.25	–	200	95
5h	CH_3	>6.25	–	500	94
5i	OCH_3	>6.25	–	50	97
5j	OC_2H_5	>6.25	–	25	98
5k	$NHCOCH_3$	>6.25	–	250	95
6a	H	>6.25	–	500	95
6b	Cl	>6.25	–	62.5	96
6c	Br	>6.25	–	12.5	99
6d	F	>6.25	–	50	98
6e	I	>6.25	–	100	97
6f	NO_2	>6.25	–	250	96
6g	CN	>6.25	–	100	98
6h	CH_3	>6.25	–	200	95
6i	OCH_3	>6.25	–	25	98
6j	OC_2H_5	>6.25	–	50	97
6k	$NHCOCH_3$	>6.25	–	100	96
	Isoniazid	0.20	99		
	Rifampicin	0.25	99		
	Ethambutol	3.12	99		
	Pyrazinamide	6.25	99		

[a] Each value is the mean of three independent experiment

linked to the 1,3,4-oxadiazole core via sulphur linkage showed higher mycobacterial inhibitory efficacies (99 %) at 12.5 μg/mL of MIC. The said two analogues were found to exhibit half fold activity against the mentioned mycobacterial strain when compared to that of standard drug pyrazinamide (6.25 μg/mL). In addition, analogue **5d** with fluorine atom and **5j** with ethoxy functional group insertion to the series involving *N*-phenyl acetamide moiety along with analogue (**6i**) from benzothiazole series bearing methoxy substituent showed appreciable activity against mycobacterial strain H37Rv at 25 μg/mL of MIC with 98–99 % of inhibition. The said profile of these analogues can be compared to that observed in the antimicrobial bioassay; however, many of the final analogues exhibited moderate activity at 50–62.5 μg/mL of MIC, whereas some were found inactive with the MIC level >100 μg/mL.

The relationship between lipophilicity (Log*P*) and MICs of final analogues was discussed, in which

(a) Among the most potent analogues tested, in case of *B. cereus*, *E. coli*, *P. aeruginosa*, *P. vulgaris*, *K. pneumoniae*, *S. typhi* bacteria and *A. clavatus*, *A. fumigatus* fungi, compounds with higher lipophilicity showed highest activity and the activity is found to decrease with the decrease in the lipophilicity value (Log*P*).
(b) In case of active analogues against *S. aureus*, *S. flexneri* bacteria and *A. niger*, *C. albicans* fungi as well as mycobacteria (H37Rv), the above-mentioned

relationship was not observed, i.e. there was a random relationship between lipophilicity of the potent analogues versus MIC figures.

(c) In case of active analogues against *A. clavatus* fungi, the opposite trend was observed with lipophilicity versus MIC values as the least lipophilic analogue exhibited highest activity and the activity was found to decrease with increase of lipophilicity of the corresponding active analogues.

Conclusion

Two series of coumarin-based 1,3,4-oxadiazol-2ylthio-*N*-phenyl(benzothiazolyl) acetamide analogues were synthesized and screened against wide range of pathogenic bacteria, fungi and mycobacteria. The bioassay results demonstrated that some analogues were potential active against all the microorganisms at MICs 3.12–25 μg/mL. Analogues with *N*-benzothiazolyl-2-chloroacetamide constituents displayed improved activity and can be highlighted as new active leads that provide a powerful incentive for further research in this area. Overall, The MIC values of these novel compounds evidenced that the presence of halogen atom(s), alkoxy and cyano substituent gave rise to a better pharmacological potency. In addition, the relationship between MIC profiles and lipophilicity of the newer analogues was also discussed. Consequently, significant antimicrobial and antituberculosis activities displayed by a novel combination system of coumarin, oxadiazole nucleus and benzothiazole rings have encouraged us to make some modifications on basic structure of obtained compounds (**6a**–**k**) to achieve more active derivatives in ongoing studies. Moreover, we believe that against the background, the present series of compounds appear to be promising for further lead optimization to obtain compounds active against several microorganisms.

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