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Short communication

Synthesis of new 4-isopropylthiazole hydrazide analogs and some derived clubbed triazole, oxadiazole ring systems – A novel class of potential antibacterial, antifungal and antitubercular agents

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ABSTRACT

In the present study a series of 4-isopropylthiazole-2-carbohydrazide analogs, derived clubbed oxadiazole–thiazole and triazole–thiazole derivatives have been synthesized and characterized by IR, ^{1}H NMR, ^{13}C NMR, elemental and mass spectral analyses. The synthesized compounds were evaluated for their preliminary in vitro antibacterial, antifungal and antitubercular activity against *Mycobacterium tuberculosis* H_{37}Rv strain by broth dilution assay method.

The synthesized compounds **7a**, **7b**, **7d** and **4** showed an antitubercular efficacy considerably greater than that of the parent 4-isopropyl-1,3-thiazole-2-carbohydrazide **1**, suggesting that the substituted 4-isopropylthiazole-2-carbohydrazide moiety plays an important role in enhancing the antitubercular properties of this class of compounds. Compounds **2c**, **3**, **4**, **6d**, **7a** and **7b** exhibited good or moderate antibacterial and antifungal activity. Compounds **4** and **7b** showed appreciable cytotoxicity at a concentration of 250 μM.

1. Introduction

Tuberculosis (TB) is a worldwide problem: about 2 million people die each year, particularly in developing countries. It is estimated that about one-third of the world population is currently infected with *Mycobacterium tuberculosis* in its latent form and addition of nine million new cases per year threatens efforts to control the disease [1].

In addition, primary and opportunistic microbial infections have increased rapidly because of immunocompromised patients (AIDS, cancer and transplants). The deadly synergy between tuberculosis (TB) and human immunodeficiency virus (HIV) had led to the containment of tubercle bacilli (macrophages and CD_4-receptor-bearing lymphocytes) and promotes the progression of recently or remotely acquired TB infection to active disease. Therefore, the development of new drugs with activity against multi drug-resistant (MDR) TB, extensively drug-resistant (XDR) TB and latent TB is a priority task, which will shorten the chemotherapy [2].

Lipophilicity is a key property that influences the ability of a drug to reach the target by transmembrane diffusion and to have a major effect on the biological activity [3]. The azole antitubercular are regarded as emerging class and thiazoles known as lipophilic analogs of imidazoles are expected to increase log *P*. And further, isopropyl group containing thiazoles has emerged as new class of potent antibacterial and antifungals, which are reported to inhibit bacteria by blocking the biosynthesis of certain bacterial lipids and/or by additional mechanisms [4,5].

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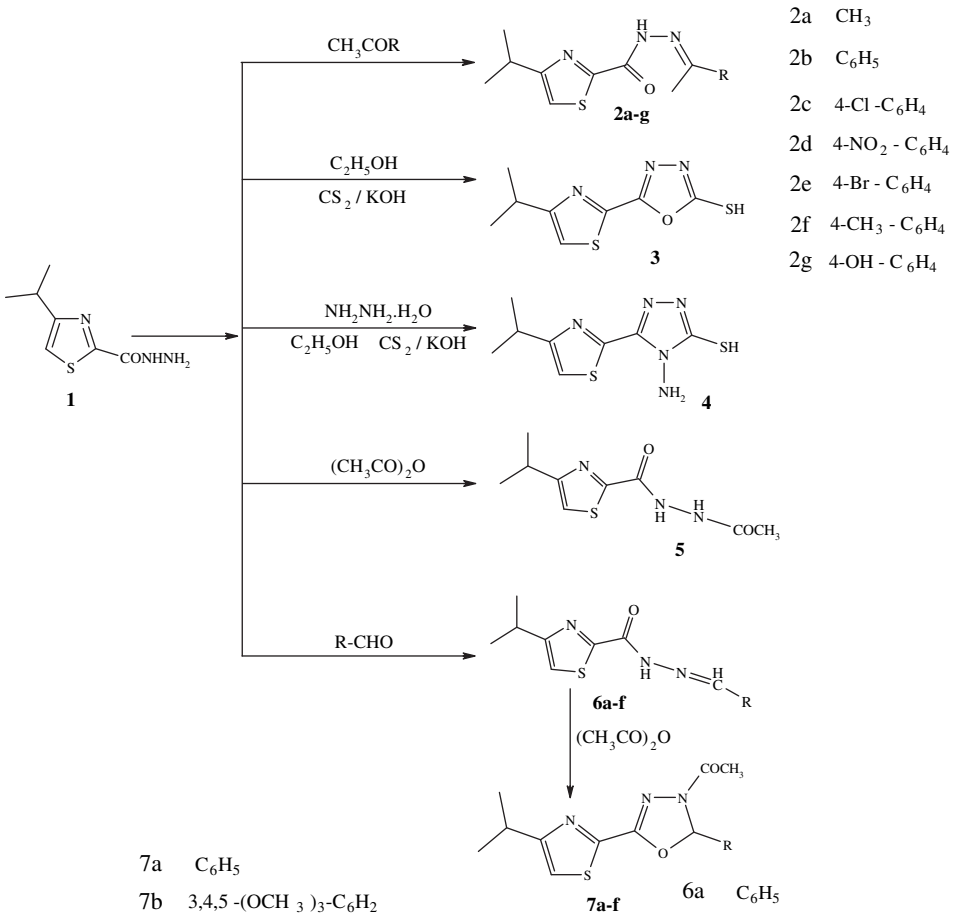
Thiazole derivatives were reported to possess antitubercular activity and their coupling with other heterocyclic rings furnishes novel biologically active compounds [6]. So after extensive literature search [7–11] and in continuation of our research work on thiazoles [12–15] it was contemplated to synthesize some novel 4-isopropylthiazoles, clubbed with 1,3,4-oxadiazole and 1,2,4-triazoles and study their antimicrobial (bacterial and fungal) activity followed by antitubercular activity.

2. Chemistry

The reaction sequences employed for synthesis of the target compounds are shown in Scheme 1, and their physical properties are depicted in Table 1. The key intermediate in the present study 4-isopropylthiazole-2-carbahydrazide **1**, was prepared by hydrazinolysis of the ethyl ester of 4-isopropylthiazole-2-carboxylate with hydrazine hydrate [16]. Reacting the starting compound **1** with acetone and appropriate substituted acetophenones resulted in the formation of corresponding hydrazone derivatives **2a**–**2g**. The preparation of 5-(4-isopropylthiazol-2-yl)-1,3,4-oxadiazole-2-thiol **3**, [17] was achieved by adopting simple one pot procedure that involves reacting compound **1** with carbon disulfide under strong basic conditions followed by acidification with dilute hydrochloric acid. Compound **1** was converted into the corresponding potassium dithiocarbazinate, which on cyclization with hydrazine hydrate yields 4-amino-5-(4-isopropyl-1,3-thiazol-2-yl)-4*H*-1,2,4-triazole-3-thiol **4**. Compound **5**, *N*-acetyl-4-isopropylthiazole-2-carbohydrazide, was synthesized by heating compound **1** with acetic anhydride. The reaction of compound **1** with different aldehydes in alcohol gave Schiff bases **6a**–**6f**, which on further heating with acetic anhydride afforded 1,3,4-oxadiazole derivatives **7a**–**7f**.

3. Biological activity

The standard strains were procured from the American Type Culture Collection (ATCC) and Gene Bank, Institute of Microbial Technology, Chandigarh, India. The antibacterial activities of the synthesized compounds were determined by broth dilution method [18,19] against the following standard bacterial strains *Staphylococcus aureus* (ATCC 11632), *Streptococcus faecalis* (ATCC 14506), *Bacillus subtilis* (ATCC 60511), *Klebsiella pneumoniae* (ATCC 10031), *Escherichia* and *Pseudomonas aeruginosa* (ATCC 10145) and



2a CH_3
2b C_6H_5
2c 4-Cl -C_6H_4
2d 4-NO_2 - C_6H_4
2e 4-Br - C_6H_4
2f 4-CH_3 - C_6H_4
2g 4-OH - C_6H_4

7a C_6H_5
7b 3,4,5 -$(OCH_3)_3$-C_6H_2
7c 4-$N(CH_3)_2$-C_6H_4
7d 4-OH-C_6H_4
7e 4-NO_2-C_6H_4
7f 2,3 -$(Cl)_2$ - C_6H_3

6a C_6H_5
6b 3,4,5 -$(OCH_3)_3$-C_6H_2
6c 4-$N(CH_3)_2$-C_6H_4
6d 4-OH-C_6H_4
6e 4-NO_2-C_6H_4
6f 2,3 -$(Cl)_2$ - C_6H_3

Scheme 1.

Table 1
Physical properties and elemental analysis data.

Compound	Molecular formula	Molecular weight	Elemental analysis (%)	
			Anal Calcd	Found
1	$C_7H_{11}N_3OS$	185.25	C – 45.39; H – 5.99; N – 22.68; S – 17.31	C – 45.78; H – 6.09; N – 22.37; S – 17.14
2a	$C_{10}H_{15}N_3O_S$	225.09	C – 53.31; H – 6.71; N – 18.65; S – 14.23	C – 54.87; H – 6.07; N – 18.23; S – 14.99
2b	$C_{15}H_{17}N_3OS$	287.11	C – 62.69; H – 5.96; N – 14.62; S – 11.16	C – 63.85; H – 5.88; N – 14.00; S – 11.56
2c	$C_{15}H_{16}ClN_3OS$	321.07	C – 55.98; H – 5.01; N – 13.06; S – 9.96	C – 55.32; H – 5.91; N – 13.76; S – 9.01
2d	$C_{15}H_{16}N_4O_3S$	332.09	C – 54.20; H – 4.85; N – 16.86; S – 9.65	C – 53.65; H – 4.59; N – 16.12; S – 9.05
2e	$C_{15}H_{16}BrN_3OS$	365.02	C – 49.19; H – 4.40; N – 11.47; S – 8.75	C – 49.06; H – 4.65; N – 11.14; S – 8.09
2f	$C_{16}H_{19}N_3OS$	301.12	C – 63.76; H – 6.35; N – 13.94; S – 10.64	C – 63.18; H – 6.11; N – 12.76; S – 10.69
2g	$C_{15}H_{17}N_3O_2S$	303.1	C – 59.38; H – 5.65; N – 13.85; S – 10.57	C – 59.09; H – 5.98; N – 13.09; S – 10.28
3	$C_8H_9N_3OS_2$	227.02	C – 42.27; H – 3.99; N – 18.49; S – 28.21	C – 42.87; H – 3.23; N – 18.43; S – 28.56
4	$C_8H_{11}N_5S_2$	241.05	C – 39.81; H – 4.59; N – 29.02; S – 26.57	C – 40.87; H – 4.03; N – 29.87; S – 26.11
5	$C_9H_{13}N_3O_2S$	227.07	C – 47.56; H – 5.77; N – 18.49; S – 14.11	C – 47.04; H – 5.05; N – 18.69; S – 14.09
6a	$C_{14}H_{15}N_3OS$	273.09	C – 61.51; H – 5.53; N – 15.37; S – 11.73	C – 61.19; H – 5.69; N – 15.30; S – 11.98
6b	$C_{17}H_{21}N_3O_4S$	363.13	C – 56.18; H – 5.82; N – 11.56; S – 8.82	C – 56.18; H – 5.82; N – 11.56; S – 8.17
6c	$C_{16}H_{20}N_4OS$	316.14	C – 60.73; H – 6.37; N – 17.71; S – 10.13	C – 60.47; H – 6.60; N – 17.02; S – 10.17
6d	$C_{14}H_{15}N_3O_2S$	289.09	C – 58.11; H – 5.23; N – 14.52; S – 11.08	C – 58.01; H – 4.87; N – 14.03; S – 11.02
6e	$C_{14}H_{14}N_4O_3S$	318.08	C – 52.82; H – 4.43; N – 17.60; S – 10.07	C – 52.56; H – 4.02; N – 17.03; S – 10.10
6f	$C_{14}H_{13}ClN_3OS$	341.02	C – 49.13; H – 3.83; N – 12.28; S – 9.37	C – 49.59; H – 3.34; N – 12.58; S – 10.48
7a	$C_{16}H_{17}N_3O_2S$	315.1	C – 60.93; H – 5.43; N – 13.32; S – 10.17	C – 60.56; H – 5.54; N – 12.89; S – 10.19
7b	$C_{19}H_{23}N_3O_5S$	405.1	C – 56.28; H – 5.72; N – 10.36; S – 7.91	C – 56.49; H – 5.68; N – 10.11; S – 7.32
7c	$C_{18}H_{22}N_4O_2S$	358.15	C – 60.31; H – 6.19; N – 15.63; S – 8.95	C – 60.98; H – 6.68; N – 14.09; S – 7.87
7d	$C_{16}H_{17}N_3O_3S$	331.1	C – 57.99; H – 5.17; N – 12.68; S – 9.68	C – 57.07; H – 4.98; N – 12.11; S – 9.23
7e	$C_{16}H_{16}N_4O_4S$	360.09	C – 53.32; H – 4.47; N – 15.55; S – 8.90	C – 53.43; H – 4.18; N – 15.87; S – 7.87
7f	$C_{16}H_{15}ClN_3O_2S$	383.03	C – 50.01; H – 3.93; N – 10.93; S – 8.34	C – 51.87; H – 3.43; N – 10.65; S – 8.54

antifungal activity against yeasts: *Saccharomyces cerevisiae* (ATCC 9763, Sc) and *Candida tropicalis* (ATCC 1369, CT), mould: *Aspergillus niger* (ATCC 6275). MIC of compounds was determined against *M. tuberculosis* $H_{37}Rv$ strain by using broth dilution assay method.

4. Results and discussion

The IR spectrum of compound **1** illustrates broad stretching band around 3517 cm^{-1} and 3335 cm^{-1} due to NH and NH_2 and strong stretching band at 1673 cm^{-1} accounting for amide carbonyl group. ^1H NMR of compound **1** shows sharp singlets at δ 9.8 and δ 4.5, accountable for NH and NH_2 which were absent on D_2O exchange. The 6 protons of terminal isopropyl group appeared as doublet at δ 1.5 and multiplet at δ 3.2 for other isopropyl proton. Thiazole proton appeared as singlet at δ 7.4. ^{13}C NMR shows a peak at 22.01 because of 2C of terminal isopropyl group and peak of C═O at 164.10, further molecular ion peak at *m*/*z* 185.06 confirms its molecular weight.

^1H NMR of compound **2d** showed a singlet at δ 2.1 due to methyl group and doublet at δ 8.1 and δ 8.32 accountable for aromatic benzyl group. ^{13}C NMR of the compound **2d** confirms resonance of CH_3 and C═N at δ 14.9 and δ 165.25 respectively. Further, mass spectrum of compound **2d** showed a molecular ion peak *m*/*z* 332.09 which confirmed its molecular weight.

Lack of ^1H NMR resonances of NH and NH_2 in the spectrum of compound **3**, further ^{13}C NMR data of compound **3** showed a peak at δ 177.5 and δ 148.32 due to C_2 and C_5 of oxadiazole and the molecular ion base peak at *m*/*z* 227.02 confirmed the structure of compound **3**.

The IR spectrum of compound **4** displayed symmetric and asymmetric stretching bands at 3353 cm^{-1} and 3248 cm^{-1} of NH/NH_2. ^1H NMR spectra show a singlet at δ 6.01 of NH_2 and a singlet at δ 14.09 (SH or HNCS) reflecting the thiol–thione tautomeric forms [18]. ^{13}C NMR spectrum of compound **4** showed signals at 185.24 and 153.15 due to C═S of triazole and C_5 of triazole respectively. The mass spectrum of compound **4** exhibited the molecular ion peak at *m*/*z* 241.05 which confirmed the molecular weight.

^1H NMR of compound **5** shows a singlet at δ 2.1, δ 10.21 and δ 7.23 due to methyl, NH and thiazole CH respectively. ^{13}C NMR spectrum shows peaks at δ 25.3, δ 167.54 and δ 111.34 due to CH_3, C═O and thiazole CH respectively and molecular ion peak at *m*/*z* 227.07 confirmed the structure.

Hydrazones **6a**–**6f** obtained from compound **1** show carbonyl amide stretching at around 1600–1650 cm^{-1} and N–H bands in 3180–3250 cm^{-1} region. ^1H NMR of compound **6a** shows sharp singlets at δ 8.32 and δ 11.97 of N═CH and NH protons. N═CH peak in ^{13}C NMR spectrum appears at δ 147.64.

In IR spectra of **7a**–**7f**, there is vanishing of NH stretching and presence of C═O bands at around 1650 cm^{-1} designated for acetyl groups. ^1H NMR of 1-(5-(4-isopropylthiazol-2-yl)-1,3,4-oxadiazole derivatives shows singlets of O–CHR–N in oxadiazoline ring at δ 6.6 and acetyl CH_3 at δ 2.21. Further same O–CHR–N in ^{13}C NMR spectrum shows peak at δ 118.1 and N–C═O of oxadiazole at δ 144.76. Compound **7a** was confirmed by molecular ion peak at *m*/*z* 315.

The results of antimicrobial testing of the synthesized compounds against selected Gram-positive, Gram-negative bacteria, yeasts, moulds and *M. tuberculosis* $H_{37}Rv$ are illustrated in Tables 2 and 3 respectively. Compounds **3** and **4** were found to be more active than other compounds at MIC 8 and 31.25 μg/mL against all tested microorganisms except a moderate potency with *S. aureus*.

Compounds **7a**–**7d** which have 2,3-dihydro-1,3,4-oxadiazole ring were more active than the starting compounds **6a**–**6f** against most of the test organisms. Interestingly **6a**–**6f** exhibited significant inhibitory (MIC 16 and 125 μg/mL) properties against Gram-negative bacteria, moulds and yeasts than Gram-positive bacteria. This implies that compounds having inductively electron withdrawing but mesomerically electron donating substituents on phenyl group were found to be the most active compounds against the test microorganisms, particularly dichloro benzene ring on compounds **6f** and **7f** had improved antimicrobial activity than other derivatives.

The synthesized compounds showed antimicrobial activity with MIC between 16 and 250 μg/mL against *S. faecalis* and had less potency against *S. aureus*. The majority of compounds exhibited good inhibitory activity against Gram-negative bacteria, moulds and yeasts than Gram-positive bacteria.

The experimental compounds showed activity against mycobacteria with MIC values ranging from 8 to 250 μg/mL. The results

Table 2
Antimicrobial activity expressed as MIC (μg/mL).

Compound	Gram-positive organisms[a]			Gram-negative organisms[a]			Fungi[a]		
	Sa	Sf	Bs	Kp	Ec	Pa	Sc	Ct	An
1	250	125	250	250	125	125	250	500	125
2a	250	125	125	125	125	125	500	125	500
2b	250	250	250	125	125	250	500	500	250
2c	125	125	31.25	125	125	125	125	125	125
2d	250	250	250	250	250	250	500	125	125
2e	250	250	125	125	250	125	31.25	125	125
2f	250	125	250	125	250	250	125	250	500
2g	125	125	250	250	125	125	31.25	31.25	125
3	125	31.25	16	16	31.25	31.25	31.25	16	31.25
4	125	31.25	8	31.25	31.25	16	16	8	8
5	500	125	250	125	250	125	500	125	500
6a	500	125	125	125	31.25	16	62.5	62.5	31.25
6b	500	250	125	125	125	31.25	125	62.5	31.25
6c	250	125	125	125	31.25	31.25	250	125	62.5
6d	125	125	125	31.25	31.25	16	31.25	16	31.25
6e	125	250	125	125	31.25	125	125	62.5	31.25
6f	250	31.25	31.25	31.25	16	31.25	16	31.25	125
7a	125	62.5	31.25	62.5	62.5	16	31.25	16	16
7b	62.5	16	16	31.25	31.25	16	31.25	16	16
7c	125	125	31.25	31.25	16	16	31.25	31.25	31.25
7d	125	31.25	16	31.25	16	31.25	31.25	16	16
7e	125	125	250	125	250	125	500	125	31.25
7f	125	16	16	125	125	31.25	16	16	31.25
Ciprofloxacin	≤5	≤5	≤1	≤1	≤1	≤5	–	–	–
Norfloxacin	≤5	≤5	≤1	≤1	≤1	≤5	–	–	–
Flucanozole	–	–	–	–	–	–	≤1	≤1	≤1

[a] The screening organisms. **Gram-positive bacteria**: *Staphylococcus aureus* (ATCC 11632, Sa), *Streptococus faecalis* (ATCC 14506, Sf), and *Bacillus subtilis* (ATCC 60511, Bs). **Gram-negative bacteria**: *Klebsiella penumoniae* (ATCC 10031, Kp), *Escherichia coli* (ATCC 10536, Ec), and *Pseudomonas aeruginosa* (ATCC 10145, Pa). **Yeasts**: *Saccharomyces cerevisiae* (ATCC 9763, Sc) and *Candida tropicalis* (ATCC 1369, Ct), mould: *Aspergillus niger* (ATCC 6275, An).

(Table 3) imply that compounds **3**, **4**, and **7a–7d** show better activity against *M. tuberculosis* $H_{37}Rv$, predominantly **3**, **4** showed highest activity (8 μg/mL) against mycobacteria.

Compounds **4**, **7b**, and **7d** were evaluated for their cytotoxic potential using A_{549} (lung adenocarcinoma) cell lines in the presence of fetal bovine serum. Compound **7d** showed maximum cytotoxicity at a concentration of 250 μM as depicted in Fig. 1. The other compounds **4** and **7b** showed appreciable cytotoxicity of about 50% of the vehicle control at a concentration of 250 μM.

Table 3
Primary antitubercular activity.

Compound	MIC values (μg/mL) of *Mycobacterium tuberculosis* $H_{37}Rv$
1	125
2a	125
2b	125
2c	62.5
2d	125
2e	31.25
2f	125
2g	125
3	8
4	8
5	125
6a	250
6b	125
6c	125
6d	125
6e	125
6f	125
7a	31.25
7b	31.25
7c	62.5
7d	31.25
7e	62.5
7f	62.5
Isoniazid	0.25

5. Experimental

5.1. Chemical protocols

Melting points were determined in open capillary tubes in a Thomas Hoover melting point apparatus and are uncorrected. Infrared spectra were recorded on (Shimadzu FT-IR 157), ^{1}H NMR and ^{13}C NMR spectra were recorded (in $CDCl_3$/DMSO-d_6) on a Bruker spectrometer at 300/400 MHz using TMS as an internal standard. Mass spectra (EI) were recorded on (AMD-604) mass spectrometer operating at 70 eV. Elemental analysis was performed on Thermo Finnigan Flash (EA 1112 CHNS Analyzer).

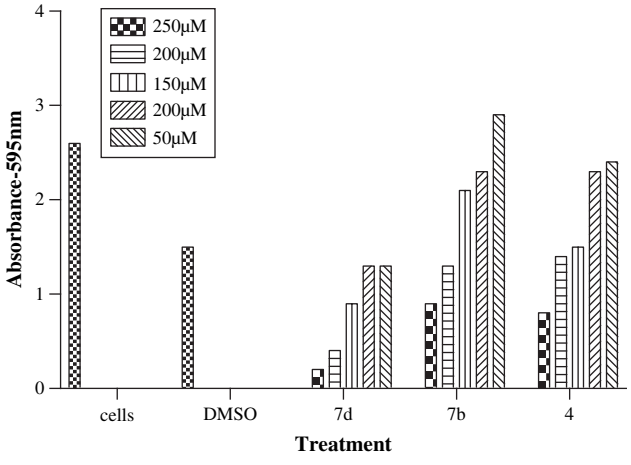


Fig. 1. Cytotoxic activity of compounds **4**, **7b** and **7d** tested in A_{549} cells by MTT assay. The bars reflect the viable cells in each treatment. Cells alone without any treatment, DMSO denote the vehicle control. The experiment was done in duplicate with triplicate readings of each experiment.

Thin layer chromatography (TLC) was performed through out the reaction to optimize the reaction for purity and completion of reaction on Merck silica gel GF_{254} aluminium sheets using mixture of different polar and nonpolar solvents in varying proportions and spots were observed using iodine as visualizing agent.

*5.1.1. Synthesis of 4-isopropyl-1,3-thiazole-2-carbohydrazide (**1**)*

Compound **1** was synthesized by refluxing a mixture of ethyl 4-isopropylthiazole-2-carboxylate (0.015 mol) with hydrazine hydrate (1.6 mL) in absolute ethanol (20 mL) for 5 h. The reaction mixture was cooled and the crystalline mass obtained was recrystallised from ethanol and obtained as yellow crystals in 84% yield.

M.p. 130–132 °C.

IR (KBr) ν max, cm^{-1}: 3517/3335 (NH/NH_2), 1673 (amide C=O).

^1H NMR (DMSO-d_6, 300 MHz) δ: 9.8 (s, 1H, NH, disappeared on D_2O exchange), 7.4 (s, 1H, thiazole C_5), 4.5 (s, 2H, NH_2, disappeared on D_2O exchange), 3.2 (m, 1H, isopropyl), 1.5 (d, $J = 8.5$ Hz, 6H, CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 168.46 (thiazole C_4), 164.10 (amide C=O), 155.21 (thiazole C_2), 119.21 (thiazole C_5), 36.11 (tertiary-1C-isopropyl), 22.01 (terminal $2CH_3$ – isopropyl) ppm.

MS (%) 185.06 (M^+ 100.0), 186.07 (7.7), 187.06 (4.6), 186.06 (1.9).

*5.1.2. General procedure for the synthesis of N-(substituted)-4-isopropyl-thiazole-2-carbohydrazide (**2a**–**2g**)*

Equimolar quantities of 4-isopropyl-1,3-thiazole-2-carbohydrazide **1** and different substituted ketones were refluxed in alcohol for 4 h in the presence of few drops of glacial acetic acid. The solvent was evaporated and the product was poured onto cold water, filtered and dried. The crude solid was recrystallised in appropriate solvent systems to give the products.

*5.1.2.1. Synthesis of 4-isopropyl-N-(propan-2-ylidene)thiazole-2-carbohydrazide (**2a**).* Recrystallised from ethanol and obtained as pale yellow crystals in 75% yield.

M.p. 208–210 °C.

IR (KBr) ν max, cm^{-1}: 3251 (NH), 1623 (amide C=O).

^1H NMR (DMSO-d_6, 300 MHz) δ: 10.21 (s, 1H, NH, disappeared on D_2O exchange), 7.41 (s, 1H, thiazole C_5), 3.23 (m, 1H, isopropyl), 1.31 (d, $J = 8.5$ Hz, 6H, CH_3), 0.91 (s, 6H, CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 170.21 (amide C=O), 162.21 (thiazole C_4), 152.13 (thiazole C_2), 114.19 (thiazole C_5), 38.12 (tertiary-1C-isopropyl), 23.12 (terminal $2CH_3$ – isopropyl), 22.12 (CH_3), 15.51 (CH_3) ppm.

MS (%) 225.09 (M^+ 100.0), 226.10 (11.0), 227.09 (4.7), 226.09 (1.9).

*5.1.2.2. Synthesis of 4-isopropyl-N-(phenylethylidene)thiazole-2-carbohydrazide (**2b**).* Recrystallised from aqueous DMF as yellow crystals in 65% yield.

M.p. 234–236 °C.

IR (KBr) ν max, cm^{-1}: 3302 (NH), 1639 (amide C=O).

^1H NMR (DMSO-d_6, 300 MHz) δ: 10.69 (s, 1H, NH, disappeared on D_2O exchange), 7.24 (s, 1H, thiazole C_5), 3.21 (m, 1H, isopropyl), 1.24 (d, $J = 8.5$ Hz, 6H, CH_3), 0.93 (s, 3H, CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 168.27 (amide C=O), 162.21 (thiazole C_4), 131.23 (C_4 – phenyl), 134.23 (C_1 – phenyl), 129.14 (C_2 and C_6), 128.53 (C_3 and C_5), 114.19 (thiazole C_5), 38.13 (tertiary-1C-isopropyl), 23.12 (terminal $2CH_3$ – isopropyl), 19.5 (CH_3) ppm.

MS (%) 287.11 (M^+ 100.0), 288.11 (18.2), 289.11 (5.0), 289.12 (1.3).

*5.1.2.3. Synthesis of 4-isopropyl-N-(1-(4-chlorophenyl)ethylidene)thiazole-2-carbohydrazide (**2c**).* Recrystallised from ethanol as yellow crystals in 61% yield.

M.p. 183–186 °C.

IR (KBr) ν max, cm^{-1}: 3323 (NH), 1636 (amide C=O).

^1H NMR (DMSO-d_6, 300 MHz) δ: 10.21 (s, 1H, NH, disappeared on D_2O exchange), 7.61 (d, $J = 8.5$ Hz, 2H, phenyl), 7.34 (s, 1H, thiazole C_5), 7.24 (d, $J = 8.5$ Hz, 2H, phenyl), 3.25 (m, 1H, isopropyl), 1.23 (d, $J = 8.5$ Hz, 6H, CH_3), 0.9 (s, 3H, CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 174.13 (amide C=O), 162.86 (thiazole C_4), 154.43 (thiazole C_2), 137.84 (C_4 – phenyl), 134.53 (C_2 and C_6), 130.71 (C_3 and C_5), 130.35 (C_1 – phenyl), 111.36 (thiazole C_5), 37.12 (tertiary-1C-isopropyl), 23.65 (terminal $2CH_3$ – isopropyl), 19.54 (CH_3) ppm.

MS (%) 321.07 (M^+ 100.0), 323.07 (37.0), 322.07 (18.2), 324.07 (6.3), 325.06 (1.4), 323.08 (1.3).

*5.1.2.4. Synthesis of 4-isopropyl-N-(1-(4-nitrophenyl)ethylidene)thiazole-2-carbohydrazide (**2d**).* Recrystallised from ethanol as yellow crystals in 81% yield.

M.p. 194–198 °C.

IR (KBr) ν max, cm^{-1}: 3322 (NH), 1598 (amide C=O).

^1H NMR (DMSO-d_6, 300 MHz) δ: 10.91 (s, 1H, NH, disappeared on D_2O exchange), 8.32 (d, 2H, phenyl), 8.12 (d, 2H, phenyl), 7.49 (s, 1H, thiazole C_5), 3.18 (m, 1H, isopropyl), 2.1 (s, 3H, CH_3), 1.35 (d, $J = 8.5$ Hz, 6H, CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 165.25 (amide C=N), 160.95 (thiazole C_4), 155.23 (thiazole C_2), 154.32 (C_4 – phenyl), 145.53 (C_1 – phenyl), 127.5 (C_2 and C_6), 119.32 (thiazole C_5), 116.76 (C_3 and C_5), 31.07 (tertiary-1C-isopropyl), 22.32 (terminal $2CH_3$ – isopropyl), 14.94 (CH_3) ppm.

MS (%) 332.09 (M^+ 100.0), 333.10 (16.5), 334.09 (4.8), 333.09 (2.3), 334.10 (2.0).

*5.1.2.5. Synthesis of 4-isopropyl-N-(1-(4-bromophenyl)ethylidene)-thiazole-2-carbohydrazide (**2e**).* Recrystallised from ethanol as yellow crystals in 66% yield.

M.p. 196–199 °C.

IR (KBr) ν max, cm^{-1}: 3341 (NH), 1643 (amide C=O).

^1H NMR (DMSO-d_6, 300 MHz) δ: 10.19 (s, 1H, NH, disappeared on D_2O exchange), 7.65 (s, 1H, thiazole C_5), 7.51 (m, 4H, phenyl), 3.43 (m, 1H, isopropyl), 1.23 (d, $J = 8.5$ Hz, 6H, CH_3), 0.9 (s, 3H, CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 176.32 (amide C=O), 162.43 (thiazole C_4), 155.34 (thiazole C_2), 133.32 (C_1 – phenyl), 131.11 (C_2 and C_6), 129.08 (C_3 and C_5), 122.22 (C_4 – phenyl), 112.56 (thiazole C_5), 38.09 (tertiary-1C-isopropyl), 23.32 (terminal $2CH_3$ – isopropyl), 19.07 (CH_3) ppm.

MS (%) 367.02 (M^+ 100.0), 365.02 (97.7), 366.02 (17.8), 368.02 (17.1), 369.01 (4.3), 369.02 (1.7), 367.03 (1.2), 368.01 (1.1).

*5.1.2.6. Synthesis of 4-isopropyl-N-(1-*p*-tolylethylidene)thiazole-2-carbohydrazide (**2f**).* Recrystallised from ethanol as yellow crystals in 60% yield.

M.p. 185–188 °C.

IR (KBr) ν max, cm^{-1}: 3332 (NH), 1621 (amide C=O).

^1H NMR (DMSO-d_6, 300 MHz) δ: 10.54 (s, 1H, NH, disappeared on D_2O exchange), 7.51 (d, 2H, phenyl), 7.23 (s, 1H, thiazole C_5), 7.13 (d, 2H, phenyl), 3.21 (m, 1H, isopropyl), 1.38 (d, $J = 8.5$ Hz, 6H, CH_3), 2.12 (s, 3H, CH_3), 0.94 (s, 3H, CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 170.11 (amide C=O), 161.19 (thiazole C_4), 157.32 (thiazole C_2), 143.1(C_4 – phenyl), 129.00 ($C_{2,3,5,6}$ – phenyl), 114.21 (thiazole C_5), 38.11 (tertiary-1C-isopropyl), 24.11 (CH_3), 23.47 (terminal $2CH_3$ – isopropyl), 19.13 (CH_3) ppm.

MS (%) 301.12 (M^+ 100.0), 302.13 (17.6), 303.12 (4.5), 303.13 (2.0), 302.12 (1.9).

*5.1.2.7. Synthesis of 4-isopropyl-N-(1-(4-hydroxyphenyl)ethylidene)thiazole-2-carbohydrazide (**2g**).* Recrystallised from ethanol as yellow crystals in 56% yield.

M.p. 176–178 °C.

IR (KBr) ν max, cm^{-1}: 3494 (OH), 3329 (NH), 1613 (amide C=O).

^1H NMR (DMSO-d_6, 300 MHz) δ: 10.23 (s, 1H, NH, disappeared on D_2O exchange), 7.12 (d, 2H, phenyl), 7.11 (s, 1H, thiazole C_5), 6.02 (d, 2H, phenyl), 5.11 (s, 1H, OH), 3.21 (m, 1H, isopropyl), 1.32 (d, $J = 8.5$ Hz, 6H, CH_3), 0.91 (s, 3H, CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 170.54 (amide C=O), 164.54 (C_4 – phenyl), 162.34 (thiazole C_4), 153.45 (thiazole C_2), 130.08 (C_2 and C_6), 119.66 (C_3 and C_5), 114.87 (thiazole C_5), 38.97 (tertiary-1C-isopropyl), 23.11 (terminal $2CH_3$ – isopropyl), 19.13 (CH_3) ppm.

MS (%) 303.10 (M^+ 100.0), 304.11 (16.5), 305.10 (4.7), 304.10 (1.9), 305.11 (1.8).

5.1.3. Synthesis of 5-(4-isopropylthiazol-2-yl)-1,3,4-oxadiazole-2-thiol (**3**)

Acid hydrazide **1** (0.003 mol) was dissolved in a solution of potassium hydroxide (0.006 mol) in water (2 mL) and ethanol (20 mL). Carbon disulfide (2 mL) was then added while stirring and the reaction mixture was heated under reflux for 8 h. The solvents were removed under reduced pressure; the residue was treated with water and then filtered. The filtrate was cooled, neutralized to pH 6 using dilute hydrochloric acid and the separated product was filtered, washed with water, dried and recrystallised from benzene as yellow crystals in 86% yield.

M.p. 208–210 °C.

IR (KBr) ν max, cm^{-1}: 2765 (SH), 1670 (C=N).

^1H NMR (DMSO-d_6, 300 MHz) δ: 14.31 (s, 1H, SH), 7.41 (s, 1H, thiazole C_5), 3.21 (m, 1H, isopropyl), 1.24 (d, $J = 8.5$ Hz, 6H, CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 177.5 (oxadiazole C_2), 162.49 (thiazole C_4), 154.45 (thiazole C_2), 148.32 (oxadiazole C_5), 112.33 (thiazole C_5), 39.04 (tertiary-1C-isopropyl), 23.45 (terminal $2CH_3$ – isopropyl) ppm.

MS (%) 227.02 (M^+ 100.0), 228.02 (11.5), 229.01 (9.0).

5.1.4. Synthesis of 4-amino-5-(4-isopropyl-1,3-thiazol-2-yl)-4H-1,2,4-triazole-3-thiol (**4**)

To a solution of potassium hydroxide (0.0067 mol) in absolute ethanol (30 mL), 4-isopropyl-1,3-thiazole-2-carbohydrazide **1** (0.003 mol) and carbon disulphide (0.006 mol) were added and the mixture was agitated for 16 h. To the resulting solution anhydrous ether was added and the precipitated potassium dithiocarbazinate was collected by filtration, washed with ether and dried under vacuum. The potassium salt was obtained in quantitative yield and was used in the next step without further purification. A suspension of the potassium salt, hydrazine hydrate (1.5 mL) and water (1.0 mL) was heated under reflux for 5 h, hydrogen sulphide evolved and homogeneous solution resulted which was diluted with 50 mL water and subsequent acidification with dilute acetic acid gave a white precipitate which was filtered, washed with water and recrystallised from aqueous DMF and obtained as pale yellow crystals in 61% yield.

M.p. 250–252 °C.

IR (KBr) ν max, cm^{-1}: 3353 (NH), 3248 (NH_2).

^1H NMR (DMSO-d_6, 300 MHz) δ: 14.09 (s, 1H, NH–C=S), 7.11 (s, 1H, thiazole C_5), 6.01 (s, 2H, NH_2, disappeared on D_2O exchange), 3.15 (m, 1H, isopropyl), 1.23 (d, $J = 8.5$ Hz, 6H, CH_3), 0.9 (s, 6H, CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 185.24 (C=S), 162.21 (thiazole C_4), 155.45 (thiazole C_2), 153.15 (triazole C_5), 144.27 (triazole C_2), 114.19 (thiazole C_5), 38.123 (tertiary-1C-isopropyl), 23.12 (terminal $2CH_3$ – isopropyl), 22.12 (CH_3) ppm.

MS (%) 241.05 (M^+ 100.0), 243.04 (9.1), 242.05 (8.8), 242.04 (3.4), 244.04 (1.0).

5.1.5. Synthesis of N-acetyl-4-isopropylthiazole-2-carbohydrazide (**5**)

Acid hydrazide **1** (0.003 mol) was warmed with acetic anhydride (5 mL) for 1 h and then the mixture was allowed to attain room temperature. The deposited pale yellow solid was filtered, washed and recrystallised from ethanol and obtained as yellow crystals in 75% yield. M.p. 202–204 °C.

IR (KBr) ν max, cm^{-1}: 3347 (NH), 1687 (acetyl C=O), 1623 (amide C=O).

^1H NMR (DMSO-d_6, 300 MHz) δ: 10.21 (s, 1H, NH, disappeared on D_2O exchange), 9.76 (s, 1H, amide NH, disappeared on D_2O exchange), 7.23 (s, 1H, thiazole C_5), 3.15 (m, 1H, isopropyl), 2.14 (s, 3H, CH_3), 1.29 (d, $J = 8.5$ Hz, 6H, CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 171.34 (acetyl C=O), 167.54 (amide C=O), 162.75 (thiazole C_4), 155.34 (thiazole C_2), 111.34 (thiazole C_5), 36.22 (tertiary-1C-isopropyl), 25.3 (CH_3), 23.12 (terminal $2CH_3$ – isopropyl) ppm.

MS (%) 227.07 (M^+ 100.0), 228.08 (10.0), 229.07 (4.6%), 228.07 (1.9).

5.1.6. General procedure for the synthesis of N-(arylidene)-4-isopropyl-1,3-thiazole-2-carbohydrazide (**6a–6f**)

Equimolar quantities of 4-isopropyl-1,3-thiazole-2-carbohydrazide **1** and substituted aldehydes were refluxed in alcohol for 3 h in the presence of few drops of glacial acetic acid. The solvent was evaporated and the product was poured on cold water, filtered and dried. The crude solid was recrystallised in appropriate solvent systems to give the products.

5.1.6.1. Synthesis of N-benzylidene-4-isopropylthiazole-2-carbohydrazide (**6a**).

Recrystallised from aqueous DMF as yellow crystals in 84% yield.

M.p. 198–200 °C.

IR (KBr) ν max, cm^{-1}: 3194 (NH), 1643 (amide C=O).

^1H NMR (DMSO-d_6, 300 MHz) δ: 11.97 (s, 1H, NH, disappeared on D_2O exchange), 8.32 (s, 1H, –N=CH–Ar), 7.2–7.8 (5H of phenyl), 7.28 (s, 1H, thiazole C_5), 3.24 (m, 1H, isopropyl) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 164.54 (thiazole C_4), 161.22 (amide C=O), 155.33 (thiazole C_2), 147.64 (CH=N),), 134.2 (phenyl C_1), 131.2 (phenyl C_4), 129.7 (phenyl C_2 and C_6), 126.12 (phenyl C_3 and C_5), 122.34 (thiazole C_5), 36.22 (tertiary-1C-isopropyl), 23.12 (terminal $2CH_3$ – isopropyl) ppm.

MS (%) 275.11 (M^+ 100.0), 276.11 (17.1), 277.11 (5.0%), 277.12 (1.1).

5.1.6.2. N-(3,4,5-Trimethoxybenzylidene)-4-isopropyl-1,3-thiazole-2-carbohydrazide (**6b**).

Recrystallised from aqueous DMF as yellow crystals in 81% yield.

M.p. 210–212 °C.

IR (KBr) ν max, cm^{-1}: 3180 (NH), 1616 (amide C=O).

^1H NMR (DMSO-d_6, 300 MHz) δ: 11.97 (s, 1H, NH, disappeared on D_2O exchange), 8.32 (s, 1H, –N=CH–Ar), 7.81 (d, 2H, $J = 8.5$ Hz, C_2 and C_5-H), 7.28 (s, 1H, thiazole C_5), 3.24 (m, 1H, isopropyl), 2.33 (s, 9H, OCH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 164.33 (amide C=O), 162.67 (thiazole C_4), 155.43 (thiazole C_2), 151.1 (phenyl C_3 and C_5), 143.23 (CH=N), 141.3 (phenyl C_4), 128.2 (phenyl C_1), 115.23 (thiazole C_5), 106.2 (phenyl C_2), 51.2 (3° CH_3), 38.91 (tertiary-1C-isopropyl), 23.23 (terminal $2CH_3$ – isopropyl) ppm.

MS (%) 363.13 (M^+ 100.0), 364.13 (18.8), 365.12 (4.5), 365.13 (2.8), 364.12 (1.9).

5.1.6.3. N-(4-Dimethylaminobenzylidene)-4-isopropyl-1,3-thiazole-2-carbohydrazide (**6c**).

Recrystallised from aqueous DMF as yellow crystals in 82% yield.

M.p. 194–200 °C.

IR (KBr) ν max, cm^{-1}: 3194 (NH), 1606 (amide C=O).

^1H NMR (DMSO-d_6, 300 MHz) δ: 11.65 (s, 1H, NH, disappeared on D_2O exchange), 8.11 (s, 1H, –N=CH–Ar), 7.32 (d, 2H, $J = 8.5$ Hz,

phenyl C_2 and C_6-H), 7.12 (s, 1H, thiazole C_5), 6.62 (d, 2H, phenyl C_3 and C_5-H), 3.26 (m, 1H, isopropyl), 2.87 (s, 6H, N–$(CH_3)_2$) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 164.11 (amide C=O), 163.51 (thiazole C_4), 155.43 (thiazole C_2), 141.12 (CH=N), 139.09 (phenyl C_4), 134.87 (phenyl C_3 and C_5), 130.6 (phenyl C_1), 126.09 (phenyl C_2 and C_6), 113.54 (thiazole C_5), 40.3 (N–$(CH_3)_2$), 35.21 (tertiary-1C-isopropyl), 22.12 (terminal $2CH_3$ – isopropyl) ppm.

MS (%) 316.14 (M^+ 100.0), 317.14 (18.4), 318.13 (4.5), 318.14 (2.0), 317.13 (1.5).

*5.1.6.4. N-(4-Hydroxybenzylidene)-4-isopropyl-1,3-thiazole-2-carbohydrazide (**6d**).* Recrystallised from aqueous DMF as pale yellow crystals in 72% yield.

M.p. 189–194 °C.

IR (KBr) ν max, cm^{-1}: 3483 (OH), 3232 (NH), 1621 (amide C=O).

^{1}H NMR (DMSO-d_6, 300 MHz) δ: 11.21 (s, 1H, NH, disappeared on D_2O exchange), 8.32 (s, 1H, –N=CH–Ar), 7.16 (d, 2H, $J = 8.5$ Hz, phenyl C_2 and C_6-H), 7.12 (s, 1H, thiazole C_5), 6.82 (d, 2H, phenyl C_3 and C_5-H), 5.09 (s, 1H, OH), 3.26 (m, 1H, isopropyl) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 166.87 (amide C=O), 165.87 (phenyl C_4), 163.11 (thiazole C_4), 157.34 (thiazole C_2), 141.23 (CH=N), 123.09 (phenyl C_1), 121.66 (phenyl C_2 and C_6), 113.43 (thiazole C_5), 111.1 (phenyl C_3 and C_5), 35.45 (tertiary-1C-isopropyl), 22.43 (terminal $2CH_3$ – isopropyl) ppm.

MS (%) 289.09 (M^+ 100.0), 290.09 (17.3), 291.08 (4.5), 291.10 (1.1).

*5.1.6.5. N-(4-Nitrobenzylidene)-4-isopropyl-1,3-thiazole-2-carbohydrazide (**6e**).* Recrystallised from aqueous DMF as pale yellow crystals in 66% yield.

M.p. 214–216 °C.

IR (KBr) ν max, cm^{-1}: 3226 (NH), 1632 (amide C=O).

^{1}H NMR (DMSO-d_6, 300 MHz) δ: 11.32 (s, 1H, NH, disappeared on D_2O exchange), 8.72 (d, 2H, phenyl C_3 and C_5-H), 8.21 (s, 1H, –N=CH–Ar), 7.98 (d, 2H, $J = 8.5$ Hz, phenyl C_2 and C_6-H), 7.12 (s, 1H, thiazole C_5), 3.26 (m, 1H, isopropyl) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 165.76 (amide C=O), 165.42 (thiazole C_4), 155.34 (thiazole C_2), 154.99 (phenyl C_4), 143.012 (phenyl C_1), 141.88 (CH=N), 132.54 (phenyl C_2 and C_6), 123.1 (phenyl C_3 and C_5), 111.1 (thiazole C_5), 33.56 (tertiary-1C-isopropyl), 23.65 (terminal $2CH_3$ – isopropyl) ppm.

MS (%) 318.08 (100.0), 319.08 (17.7), 320.07 (4.5), 320.09 (1.1).

*5.1.6.6. N-(2,3-Dichloro-benzylidene)- 4-isopropyl-1,3-thiazole-2-carbohydrazide (**6f**).* Recrystallised from aqueous DMF as pale yellow crystals in 66% yield.

M.p. 208–213 °C.

IR (KBr) ν max, cm^{-1}: 3216 (NH), 1632 (amide C=O).

^{1}H NMR (DMSO-d_6, 300 MHz) δ: 11.32 (s, 1H, NH, disappeared on D_2O exchange), 8.43 (s, 1H, –N=CH–Ar), 7.64 (d, H, phenyl C_2), 7.43 (d, H, phenyl C_6), 7.13 (d, H, phenyl C_5-H), 7.12 (s, 1H, thiazole C_5), 3.26 (m, 1H, isopropyl) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 171.12 (amide C=O), 162.65 (thiazole C_4), 155.68 (thiazole C_2), 143.11(CH=N), 135.1 (phenyl C_3 and C_4), 133.09 (phenyl C_1), 132.54 (phenyl C_2), 124.32 (phenyl C_5), 114.43 (thiazole C_5), 38.96 (tertiary-1C-isopropyl), 23.11 (terminal $2CH_3$ – isopropyl) ppm.

MS (%) 341.02 (M^+ 100.0), 343.01 (68.5), 342.02 (16.1), 345.01 (13.2), 344.02 (9.9), 346.01 (2.2), 344.01 (2.0), 343.02 (1.6), 342.01 (1.1).

*5.1.7. General procedure for the synthesis of 3-acetyl-5-(4-isopropylthiazole)-2-substituted-2,3-dihydro-1,3,4-oxadiazoles (**7a**–**7f**)*

A mixture of **6a**–**6f** (0.003 mol) and acetic anhydride (10 mL) was heated under reflux for 4 h. After the reaction mixture attained room temperature, excess acetic anhydride was decomposed by water and the mixture was stirred for further 30 min. The separated product was filtered, washed with water, dried and recrystallised in appropriate solvent systems to give the products.

*5.1.7.1. 1-(5-(4-Isopropylthiazol-2-yl)-2-phenyl-1,3,4-oxadiazol-3(2H)-yl)ethanone (**7a**).* Recrystallised from ethanol as pale brown crystals in 60% yield.

M.p. 154–156 °C.

IR (KBr) ν max, cm^{-1}: 1665 (amide C=O).

^{1}H NMR (DMSO-d_6, 300 MHz) δ: 7.32 (s, 1H, thiazole C_5), 7.23 (d, 5H, Ar-H), 6.64 (s, oxadiazole C_2-H), 3.26 (m, 1H, isopropyl), 2.21 (s, 3H, CH_3), 1.23 (d, 6H, terminal CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 170.11 (acetyl C=O), 160.98 (thiazole C_4), 155.69 (thiazole C_2), 144.76 (oxadiazole C_5), 128.11 (phenyl C_4), 125.98 (phenyl C_3 and C_5), 123.11 (phenyl C_2,C_6), 111.09 (thiazole C_5), 76.21 (oxadiazole C_2), 38.12 (tertiary-1C-isopropyl), 23.54 (acetyl CH_3), 23.24 (terminal $2CH_3$ – isopropyl) ppm.

MS (%) 315.10 (M^+ 100.0), 316.11 (17.6), 317.10 (4.7), 317.11 (2.0), 316.10 (1.9).

*5.1.7.2. 1-(5-(4-Isopropylthiazol-2-yl)-2-(3,4,5-trimethoxyphenyl)-1,3,4-oxadiazol-3(2H)-yl)ethanone (**7b**).* Recrystallised from ethanol as pale brown crystals in 67% yield.

M.p. 188–192 °C.

IR (KBr) ν max, cm^{-1}: 1650 (amide C=O).

^{1}H NMR (DMSO-d_6, 300 MHz) δ: 7.01 (s, 1H, thiazole C_5), 6.63 (s, oxadiazole C_2-H), 6.12 (d, 2H, Ar-H), 4.01 (s, 9H, OCH_3), 3.12 (m, 1H, isopropyl), 2.21 (s, 3H, CH_3), 1.29 (d, 6H, terminal CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 168.11 (acetyl C=O), 162.11 (thiazole C_4), 154.11 (oxadiazole C_5), 152.01 (thiazole C_2), 151.11 (phenyl C_3,C_5), 138.01 (phenyl C_4), 134.34 (phenyl C_1), 116.76 (thiazole C_5), 103.32 (phenyl C_2,C_6), 76.36 (oxadiazole C_2), 56.6 (3C, OCH_3) 38.91 (tertiary-1C-isopropyl), 23.41 (terminal $2CH_3$ – isopropyl), 23.21 (acetyl CH_3) ppm.

MS (%) 405.14 (100.0), 406.14 (21.8), 407.13 (4.5), 407.14 (3.5), 406.13 (1.1).

*5.1.7.3. 1-(5-(4-Isopropylthiazol-2-yl)-2-(4-dimethylaminophenyl)-1,3,4-oxadiazol-3(2H)-yl)ethanone (**7c**).* Recrystallised from ethanol as pale brown crystals in 62% yield.

M.p. 182–185 °C.

IR (KBr) ν max, cm^{-1}: 1634 (amide C=O).

^{1}H NMR (DMSO-d_6, 300 MHz) δ: 7.16 (s, 1H, thiazole C_5), 7.01 (s, 2H, Ar-H), 6.67 (m, 2H, Ar-H), 6.34 (s, oxadiazole C_2-H), 3.32 (m, 1H, isopropyl), 2.91 (s, 6H, N–$(CH_3)_2$), 2.21 (s, 3H, CH_3), 1.43 (d, 6H, terminal CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 166.34 (acetyl C=O), 162.23 (thiazole C_4), 159.65 (oxadiazole C_5), 152.12 (thiazole C_2), 148.98 (phenyl C_4), 129.34 (phenyl C_1), 128.11 (phenyl C_2, C_6), 116.76 (thiazole C_5), 115.36 (phenyl C_3, C_5), 78.87 (oxadiazole C_2), 41.6 (s, 2C, N–$(CH_3)_2$), 38.41 (tertiary-1C-isopropyl), 23.33 (acetyl CH_3), 23.11 (terminal $2CH_3$ – isopropyl) ppm.

MS (%) 358.15 (M^+ 100.0), 359.15 (20.6), 360.14 (4.5), 360.15 (2.7), 359.14 (1.5), 361.15 (1.0).

*5.1.7.4. 1-(5-(4-Isopropylthiazol-2-yl)-2-(4-hydroxyphenyl)-1,3,4-oxadiazol-3(2H)-yl)ethanone (**7d**).* Recrystallised from ethanol as pale brown crystals in 60% yield.

M.p. 178–182 °C.

IR (KBr) ν max, cm^{-1}: 3503 (OH), 1632 (amide C=O).

^{1}H NMR (DMSO-d_6, 300 MHz) δ: 7.32 (s, 1H, thiazole C_5), 7.11 (m, 2H, Ar-H), 6.64 (s, oxadiazole C_2-H), 6.62 (m, 2H, Ar-H), 6.11 (s, 1H, OH), 3.45 (m, 1H, isopropyl), 2.21 (s, 3H, CH_3), 1.23 (d, 6H, terminal CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 166.98 (acetyl C=O), 162.21 (thiazole C_4), 158.12 (oxadiazole C_5), 157.21 (phenyl C_4), 152.11

(thiazole C_2), 131.39 (phenyl C_1), 128.76 (phenyl C_2, C_6), 116.65 (thiazole C_5), 112.43 (phenyl C_3, C_5), 75.76 (oxadiazole C_2), 38.54 (tertiary-1C-isopropyl), 23.87 (terminal $2CH_3$ – isopropyl), 23.34 (acetyl CH_3) ppm.

MS (%) 331.10 (M^+ 100.0), 332.10 (19.3), 333.09 (4.5), 333.11 (1.5).

*5.1.7.5. 1-(5-(4-Isopropylthiazol-2-yl)-2-(4-nitrophenyl)-1,3,4-oxadiazol-3(2H)-yl)ethanone (**7e**).* Recrystallised from ethanol as pale brown crystals in 77% yield.

M.p. 168–172 °C.

IR (KBr) ν max, cm^{-1}: 1650 (amide C=O).

^{1}H NMR (DMSO-d_6, 300 MHz) δ: 8.32 (m, 2H, Ar-H), 7.87 (s, 1H, thiazole C_5), 7.32 (m, 2H, Ar-H), 6.61 (s, oxadiazole C_2-H), 3.32 (m, 1H, isopropyl), 2.21 (s, 3H, CH_3), 1.13 (d, 6H, terminal CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 165.87 (thiazole C_4), 161.01 (acetyl C=O), 152.11 (thiazole C_2), 151.34 (oxadiazole C_5), 143.01 (phenyl C_1, C_4), 129.12 (phenyl C_2, C_6), 121.01 (phenyl C_3, C_5), 114.11 (thiazole C_5), 77.65 (oxadiazole C_2), 37.11 (tertiary-1C-isopropyl), 23.11 (acetyl CH_3), 22.21 (terminal $2CH_3$ – isopropyl) ppm.

MS (%) 360.09 (M^+ 100.0), 361.09 (19.7), 362.09 (5.8), 362.10 (1.5).

*5.1.7.6. 1-(5-(4-Isopropylthiazol-2-yl)-2-(2,3-dichlorophenyl)-1,3,4-oxadiazol-3(2H)-yl)ethanone (**7f**).* Recrystallised from ethanol as pale brown crystals in 56% yield.

M.p. 155–158 °C.

IR (KBr) ν max, cm^{-1}: 1652 (amide C=O).

^{1}H NMR (DMSO-d_6, 300 MHz) δ: 7.11–7.34 (m, 3H, Ar-H), 7.21 (s, 1H, thiazole C_5), 6.62 (s, oxadiazole C_2-H), 3.65 (m, 1H, isopropyl), 2.21 (s, 3H, CH_3), 1.31 (d, 6H, terminal CH_3) ppm.

^{13}C NMR (DMSO-d_6, 300 MHz) δ: 165.28 (acetyl C=O), 161.33 (thiazole C_4), 157.43 (thiazole C_2), 154.09 (oxadiazole C_5), 144.23 (phenyl C_1), 131.01 (phenyl C_3), 128.49 (phenyl $C_{4,5}$), 126.87 (phenyl C_6), 123.12 (phenyl $C_{2,}$), 111.34 (thiazole C_5), 76.54 (oxadiazole C_2), 38.45(tertiary-1C-isopropyl), 23.35 (acetyl CH_3), 21.01(terminal $2CH_3$ – isopropyl) ppm.

MS (%) 383.03 (M^+ 100.0), 385.02 (68.5), 384.03 (18.4), 387.02 (13.5), 386.03 (12.1), 388.02 (2.5), 385.03 (2.2), 387.03 (1.4), 386.02 (1.3), 384.02 (1.1).

5.2. Biological protocol

5.2.1. Antimicrobial activity

The antimicrobial susceptibility testing was performed in vitro by broth microdilution method [19–21]. The MIC determination of the synthesized compounds was carried out in side-by-side comparison with ciprofloxacin and norfloxacin against Gram-positive bacteria (*S. aureus*, *S. faecalis*, *B. subtilis*) and Gram-negative (*K. penumoniae*, *Escherichia coli*. *P. aeruginosa*). The antifungal activity was assayed against yeasts (*C. tropicalis*, *S. cerevisiae*) and moulds (*A. niger*). The minimal inhibitory concentrations (MIC, μg/mL) were defined as the lowest concentrations of compound that completely inhibited the growth of each strain. Test compounds (10 mg) were dissolved in dimethylsulfoxide (DMSO, 1 mL), then diluted in culture medium (Mueller–Hinton Broth for bacteria and Sabouraud Liquid Medium for fungi), further progressive dilutions were done to obtain final concentrations of 1, 2, 4, 8, 16, 31.25, 62.5, 125, 250 and 500 μg/mL. DMSO never exceeded 1% v/v. The tubes were inoculated with 10^5 cfu mL^{-1} (colony forming unit/mL) and incubated at 37 °C for 24 h. The growth control consisting of media and media with DMSO at the same dilutions as used in the experiments were employed.

5.2.2. Antitubercular activity

The preliminary antitubercular screening for test compounds was obtained for *M. tuberculosis* H_{37}Rv, the MIC of each drug was determined by broth dilution assay [22–24] and is defined as the lowest concentration of drug, which inhibits ≤99% of bacterial population present at the beginning of the assay. A frozen culture in Middlebrook 7H9 broth supplemented with 10% albumin–dextrose–catalase and 0.2% glycerol was thawed and diluted in broth to 10^5 cfu mL^{-1} (colony forming unit/mL) dilutions. Each test compound was dissolved in DMSO and then diluted in broth twice at the desired concentration. The final concentration of DMSO in the assay medium was 1.3%. Each U-tube was then inoculated with 0.05 mL of standardized culture and then incubated at 37 °C for 21 days. The growth in the U-tubes was compared with visibility against positive control (without drug), negative control (without drug and inoculum) and with standard isoniazid.

5.2.3. MTT assay for cell viability

Toxicity of compounds **4**, **7b**, and **7d** in A_{549} cell lines in the presence of 10% and 0.2% FBS respectively, was determined using 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide reduction assay [25,26]. The compounds were dissolved in DMSO at 10 mM concentration and stored at −20°. The dilutions were made in culture medium before treatment.

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