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ORIGINAL RESEARCH

Synthesis and biological evaluation of 1,3,4-oxadiazole bearing dihydropyrimidines as potential antitubercular agents

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Abstract A series of 5-(4-acetyl-5-(aryl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1*H*)-ones **(4a-l)** were synthesized in very good yields (56–78 %). Biginelli adduct **1** synthesized in the first step was reacted with hydrazine hydrate to furnish carbohydrazide intermediate **2** which on further reaction with different aryl aldehydes yielded 4-(2-fluorophenyl)-6-methyl-*N*′-(aryl)-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazides **(3a-l)**. These intermediates **3a-l** were cyclized with the help of acetic anhydride to give the titled compounds **4a-l**. All the newly synthesized compounds **4a-l** were screened for their in vitro antitubercular activity against *Mycobacterium tuberculosis* $H_{37}Rv$ (Mtb) using Lowenstein–Jensen method. Compounds **4c**, **4f**, **4h**, **4i** and **4j** inhibited more than 90 % of mycobacterial growth. Minimum inhibitory concentration (MIC) for compound **4i** was found to be equipotent (MIC: 0.20 μg/ml) to the reference drug isoniazid. Structure activity relationship revealed that the presence of electron withdrawing group/atoms at *para* position of phenyl ring remarkably enhanced the antitubercular activity of synthesized compounds.

Keywords 3,4-Dihydropyrimidin-2(1*H*)-ones · 1,3,4-Oxadiazoles · Antitubercular activity: · Lowenstein–Jensen method · *Mycobacterium tuberculosis* $H_{37}Rv$

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Introduction

Tuberculosis (TB) is a highly infectious airborne disease caused by pathogenic bacterium *Mycobacterium tuberculosis* (MTB). This pathogen is responsible for over two million lives each year and dwells hidden in as many as two billion people (WHO, 2013). In addition, emergence of new virulent forms of TB such as multi-drug-resistant tuberculosis (MDR-TB) and extremely drug-resistant tuberculosis (XDR-TB) and its synergy with human immunodeficiency virus (HIV) has fueled its epidemic nature (Zumla *et al.,* 2013). There is an urgent need to develop newer, potent and safer antitubercular agents which are less prone to resistance. (Palomino and Martin, 2013; Nguyen and Jacobs, 2012; Nzila *et al.,* 2011). To cater this need, researchers around the world are actively involved in the development of antitubercular agents with improved biological properties and novel mode of action (Manvar *et al.,* 2010, 2011, 2013, 2014; Upadhyay *et al.,* 2012; Virsdoia *et al.,* 2010). One of the best tools to develop newer and effective antitubercular agents is to design hybrid molecules by molecular hybridization of different bioactive substances (Dartois and Barry, 2013; Patani and LaVoie, 1996).

3,4-Dihydropyrimidin-2(1*H*)-one (DHPM) derivatives have attracted considerable amount of attention due to the wide range of pharmacological properties such as calcium channels blockers, anticancer, antiviral, antioxidant and anti-inflammatory activity (Inca *et al,.* 2006; Prashantha Kumar *et al.,* 2009; Hélio *et al.,* 2006; Zamanova *et al.,* 2010; Sushilkumar and Devanand, 2004). Dihydropyrimidines are potential inhibitors of dihydrofolate reductase (DHFR), a promising drug target for the development of anti-infective agents. Although DHFR does not represent a novel target, there is still enthusiasm for the development

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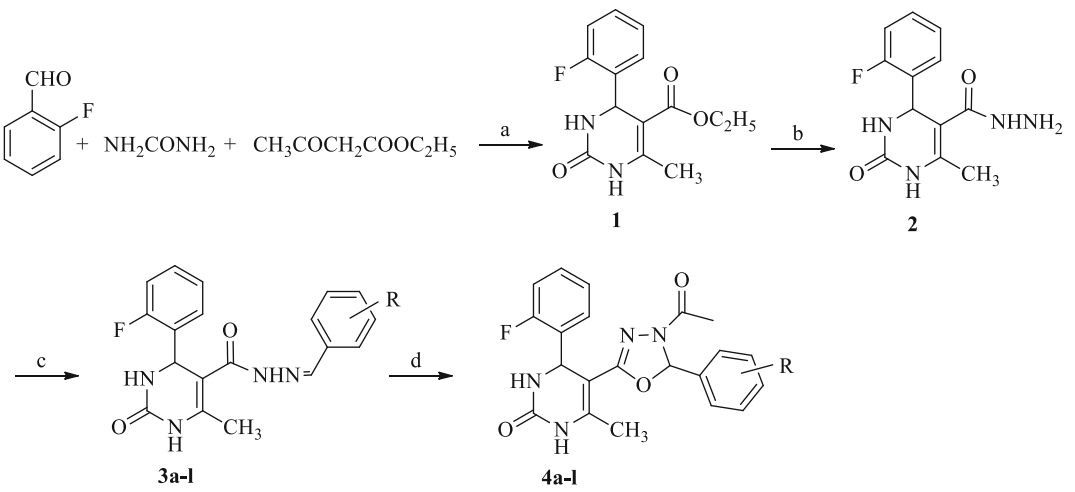
of DHFR inhibitors, particularly with regard to mycobacteria (Suling *et al.,* 1998; Li *et al.,* 2000; Suling and Maddry, 2001; Gerum *et al.,* 2002; El-Hamamsy *et al.,* 2007). On the other hand, 1,3,4-oxadiazoles are an important class of heterocyclic compounds with an array of biological and pharmacological properties such as antibacterial, antitubercular, vasodilatory, antifungal, cytotoxic, anti-inflammatory, analgesic hypolipidemic, anticancer and ulcerogenic activities (Suresh Kumar *et al.,* 2010; Shirote and Bhatia, 2011; Prakash *et al.,* 2010; Padmavathi *et al.,* 2009; Akhter *et al.,* 2009; Idrees *et al.,* 2009; Jayashankar *et al.,* 2009; Kumar *et al.,* 2009; Bhandari *et al.,* 2008). Further, 1,3,4-oxadiazole heterocycles are very good bioisosteres of amides and esters and possess a phenomenal pharmacokinetic property, lipophilicity that influences the drug's ability to reach the target by transmembrane diffusion and may demonstrate potent activity against resistant TB by inhibiting the biosynthesis of lipids (Ouellet *et al.,* 2008; Seward *et al.,* 2006; Ahsan *et al.,* 2011). Recognizing these facts and in continuation to our endeavors toward the development of anti-infective agents (Desai *et al.,* 2012a, b, 2013a, b, c, 2015), it was envisaged that the design and synthesis of such novel compounds which include advantage of dual pharmacophore of DHPMs and 1,3,4-oxadiazoles in single molecular framework are worth the attempt.

Results and discussion

Chemistry

The reaction sequence for the preparation of 5-(4-acetyl-5-(aryl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1*H*)-ones (**4a-l**) is outlined in Scheme 1. Initially, ethyl 4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carboxylate (**1**) was synthesized Biginelli reaction of urea, ethylacetoacetate and 2-fluorobenzaldehyde in methanol with catalytic amount of HCl. Compound **1** was further reacted with hydrazine hydrate in the presence of catalytic amount of con. H_2SO_4 to furnish 4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (**2**). In the next step, intermediate **2** was reacted with different aryl aldehydes in the presence of catalytic amount of glacial acetic acid to yield *N*′-arylidene-4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (**3a-l**) intermediates. Finally, these intermediates were condensed with acetic anhydride to produce the targeted compounds **4a-l**. Designed series of molecules **4a-l** were characterized by IR, ^{1}H NMR, ^{13}C NMR and mass spectrometry techniques before evaluating for in vitro antitubercular activity.

The IR spectrum of compounds **4a-l** showed the stretching vibrations of carbonyl group around 1729–1767 cm^{-1} and the absorption bands of –NH group around 3210–3267 cm^{-1}. Strongly intense absorption bands were observed around 1538–1594, 1529–1567 and 1136–1178 cm^{-1} due to stretching vibrations of –C=N, –C=C and –C–F groups, respectively. In ^{1}H NMR spectra of **4a-l**, characteristic signal of secondary amine showed a broad singlet around 5.67–6.14 ppm exchangeable with D_2O. The intense singlet peak around 2.03–2.35 ppm corresponded to the methyl protons of the free –$COCH_3$ group in 1,3,4-oxadiazole ring. Aromatic and –CH=N protons appeared in the region at 6.99–8.10 ppm. The singlet around 6.87–6.99 ppm assigned to –CH proton further confirmed the formation of 1,3,4-oxadiazole ring. ^{13}C NMR spectrum of



For **3a-l** and **4a-l**, R = -H, -2-F, -4-F, -2-OH, -4-OH, -4-Cl, -2-NO_2, -3-NO, -4-NO_2, -2,6(Cl)$_2$, -2-OCH_3, -4-OCH_3

Scheme 1 Synthetic protocol for the title compounds. Reagents and condition: *a* HC1, MeOH, Reflux; 2h; *b* NH_2NH_2 H_2O, H_2SO_4, 1,4-dioxane, Reflux, 3h; *c* Aryl aldehydes, EtOH, AcOH, Reflux; 5–6 h; *d* $(CH_3CO)_2O$, Reflux, 2h

compounds **4a-l** displayed characteristic signal of carbonyl carbon in 1,3,4-oxadiazole ring around 158.2–158.9 ppm. Carbons of –C=N in oxadiazole ring showed a chemical shift around 160.9–161.7 ppm. Chemical shift around 95.6–101.0 ppm was assigned to a –CH carbon in the oxadiazole ring. Aromatic carbons were observed in the range of 114.0–142.5 ppm. In mass spectra, molecular ion peak is in agreement with proposed molecular weight and elemental analysis.

Antimycobacterial activity

Compounds **4a-l** were initially screened for their in vitro antimycobacterial activity against *M. tuberculosis* $H_{37}Rv$ strain using Lowenstein–Jensen method exactly as described previously (Kathrotiya and Patel 2013). Results of the antitubercular studies are given in Table 1. Compounds exhibiting ≥90 % inhibition in the initial screen were retested at lower concentration (MIC) in Lowenstein–Jensen medium to determine the actual MIC. In the preliminary screening, compounds **4c**, **4f**, **4h, 4i** and **4j** inhibited MTB with 90–100. In the secondary level, compounds **4c** and **4f** inhibited MTB with MIC of 3.12 μg/mL and compound **4i** with MIC of 0.20 μg/mL. Among all these compounds, compound **4i** having 4-NO_2 substituent at the phenyl ring of 1,3,4-oxadiazole substitution was found to be the most potent compound of the series with MIC equivalent to the standard drug isoniazid. Preliminary in vitro results provide an excellent lead for further development of these molecules as novel antitubercular agents. It is interesting to note that substituents with electronic withdrawing group/atoms such as nitro, chloro and fluoro at *para* position of 1,3,4-oxadiazolyl phenyl ring demonstrated high inhibitory activity against MTB, indicating that the electronic properties of the substituents have major influence on the antimycobacterial activity. It is a well-known fact that strong electron withdrawing substitution such as nitro, chloro and fluoro at the *para* position of the aromatic ring increases the overall lipophilicity of molecule and hence facilitates diffusion of a molecule through the biological membranes to reach its site of action, which in turn may provide a positive influence on antitubercular activity (Jorge *et al.* 2009; Testa *et al.* 2000).

Table 1 In vitro antitubercular activity of **4a-l** against *M. tuberculosis* $H_{37}Rv$

Entry	% Inhibition	MIC (μg/mL)
4a	36	–
4b	62	–
4c	98	3.12
4d	69	–
4e	71	–
4f	99	3.12
4 g	56	–
4 h	99	12.5
4i	99	0.20
4j	98	100
4 k	39	–
4 l	48	–
Isoniazid	99	0.20

Experimental

All reactions except those in aqueous media were carried out by standard techniques for the exclusion of moisture. Melting points were determined on an electro-thermal melting point apparatus by open capillary method and were reported uncorrected. TLC on silica gel plates (Merck, 60, F_{254}) was used for monitoring of the reactions. Column chromatography on silica gel (Merck, 70–230 mesh and 230–400 mesh ASTH for flash chromatography) was applied when necessary to isolate and purify the reaction products. Yields refer to purified products and are not optimized. Elemental analysis (% C, H, N) was carried out by a Perkin-Elmer 2400 CHN analyzer. IR spectra of all compounds were recorded on a Perkin-Elmer FT-IR spectrophotometer in KBr, and frequencies are reported in cm^{-1}. ^1H NMR spectra were run on Varian Gemini 400 MHz and ^{13}C NMR spectra on Varian Mercury-400, 100 MHz in DMSO-d_6 as a solvent and tetramethylsilane (TMS) as an internal standard. Chemical shifts are reported as units (ppm) values. LCMS spectra were scanned on a Shimadzu LCMS 2010 spectrometer. Anhydrous reactions were carried out in oven-dried glassware in nitrogen atmosphere.

Preparation of ethyl 4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carboxylate (1)

Compound ethyl 4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carboxylate **(1)** was prepared according to the literature method with some minor modifications. Urea (0.5 mol), ethylacetoacetate (0.75 mol) and 2-fluorobenzaldehyde (0.75 mol) were mixed in methanol (25 ml). Catalytic amount of HCl was added to the reaction mixture and refluxed for 2 h. The progress of reaction was monitored by TLC. After completion of reaction, the reaction mass was cooled and white/yellowish crystals were separated. Almost pure product obtained as white/cream solid was filtered and dried. It was further crystallized using methanol. Yield: 85 %; mp 220–221 °C; Anal. calcd. for $C_{14}H_{15}N_2O_3$: C-60.42, H-5.43, N-10.07; Found: C-60.20, H-5.55, N-10.12.

Preparation of 4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-i (2)

Compound **(1)** (0.01 mol) dissolved in 1,4-dioxane (20 mL), and to this, hydrazine hydrate (99 %) (0.01 mol) was added followed by the addition of a catalytic amount of conc. H_2SO_4 and allowed to stir for 3 h at 100 °C. After completion of reaction, crude mass was allowed to cool and poured on crushed ice. Product obtained as yellowish precipitate was filtered and dried. Purification was done by crystallization using ethanol (95 %).

4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (2)

Light yellow solid; Yield: 69 %; mp 198–199 °C; IR (KBr) ν_{max} 3452, 3342, 3071, 1513, 1685 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): $\delta = 1.97$ (s, 2H, NH_2, D_2O exch.), 2.35 (s, 3H, $–CH_3$), 5.60 (s, 1H, –CH), 5.87 (s, 1H, –$NHNH_2$), 6.05 (s, 1H, –NHCPh), 6.18 (s, 1H, –$NHCCH_3$), 6.94 (d, 2H, Ar–H), 7.23 (d, 2H, Ar–H); ^{13}C NMR (DMSO-d_6, 100 MHz): $\delta = 162.5$ (–CONH–), 161.5 (–C–F), 155.9 (–C–CH_3), 150.8 (–NHN=CH–), 114.6–132.4 (Ar–C), 101.2 (–C=C–CH_3 pyrimidine ring), 52.9 (–CH pyrimidine), 16.2 ($–CH_3$); LCMS: *m/z* 262 $[M]^+$; Anal. calcd. for $C_{12}H_{13}FN_4O_2$: C, 54.54; H, 4.96; N, 21.20. Found: C, 54.92; H, 4.55; N, 21.12.

General procedure for the preparation of N′-benzylidene-4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazides (3a-l)

A mixture of different aldehydes (0.01 mol) and 4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (0.01 mol) **(2)** was dissolved in ethanol. A catalytic amount of glacial acetic acid was added into the reaction mixture and refluxed for 5–6 h at 78 °C by using reflux condenser equipped with magnetic stirrer. After completion of reaction, crude mass was cooled to room temperature and the crystals formed were filtered off and recrystallized from alcohol (95 %) to give product **(3a-l)**.

N′-benzylidene-4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (3a)

Light brown crystal; Yield: 72 %; mp 240–241 °C; IR (KBr) ν_{max} 3254, 3052, 2975, 1759, 1544, 1529, 1137 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): $\delta = 2.42$ (s, 3H, $–CH_3$), 5.52 (s, 1H, –CH pyrimidine ring), 6.05 (s, 1H, –NH–C–Ph), 7.04–7.49 (m, 9H, Ar–H), 7.46 (s, 1H, –NH–N=CH), 7.55 (s, 1H, –NH–N=CH–), 8.21 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): $\delta = 16.8$ ($–CH_3$), 51.7 (–CH pyrimidine), 99.4 (–C=C–CH_3 pyrimidine ring), 116.6 (–C=C–F in phenyl ring), 125.3 (Ar–C), 127.0 (Ar–C) (2), 128.4 (Ar–C), 129.3 (2) (Ar–C), 130.4 (Ar–C), 130.7 (Ar–C), 130.9 (Ar–C), 134.4 (Ar–C), 149.4 (–NHN=CH–), 155.3 (–CO pyrimidine ring), 155.7 (–C–CH_3), 161.3 (–C–F), 163.4 (–CONH–); LCMS: *m/z* 352 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

N′-(2-fluorobenzylidene)-4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (3b)

Light yellow crystal; Yield: 76 %; mp 179–180 °C; IR (KBr) ν_{max} 3248, 3060, 2964, 1743, 1547, 1535, 1140 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): $\delta = 2.36$ (s, 3H, $–CH_3$), 5.59 (s, 1H, –CH pyrimidine ring), 6.15 (s, 1H, –NH–C–Ph), 7.06–7.47 (m, 8H, Ar–H), 7.48 (s, 1H, –NH–N=CH), 7.54 (s, 1H, –NH–N=CH–), 8.45 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): $\delta = 16.4$ ($–CH_3$), 51.5 (–CH pyrimidine), 99.7 (–C=C–CH_3 pyrimidine ring), 115.3 (2), 116.8 (–C=C–F in phenyl ring), 125.1 (Ar–C), 130.7 (Ar–C), 130.9 (Ar–C), 131.4 (Ar–C), 131.7 (Ar–C), 132.4 (Ar–C), 132.7 (Ar–C), 149.7 (–NHN=CH–), 155.7 (–CO pyrimidine ring), 156.4 (–C–CH_3), 161.7 (–C–F), 161.9 (–C–F), 163.7 (–CONH–); LCMS *m/z* 370 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

N′-(4-fluorobenzylidene)-4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (3c)

Light orange crystal; Yield: 69 %; mp 212–213 °C; IR (KBr) ν_{max} 3247, 3065, 2982, 1747, 1560, 1533, 1145 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): $\delta = 2.42$ (s, 3H, $–CH_3$), 5.65 (s, 1H, –CH pyrimidine ring), 6.23 (s, 1H, –NH–C–Ph), 7.12–7.54 (m, 8H, Ar–H), 7.56 (s, 1H, –NH–N=CH), 7.64 (s, 1H, –NH–N=CH–), 8.37 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz):$\delta = 16.7$ ($–CH_3$), 51.8 (–CH pyrimidine), 100.4 (–C=C–CH_3 pyrimidine ring), 114.9 (2), 116.4 (–C=C–F in phenyl ring), 125.7 (Ar–C), 130.4 (Ar–C), 130.8 (Ar–C), 131.6 (Ar–C), 131.9 (Ar–C), 132.5 (Ar–C), 132.9 (Ar–C), 149.9 (–NHN=CH–), 155.8 (–CO pyrimidine ring), 156.8 (–C–CH_3), 161.9 (–C–F), 162.5 (–C–F), 164.4 (–CONH–); LCMS *m/z* 370 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

4-(2-fluorophenyl)-N′-(2-hydroxybenzylidene)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (3d)

Light gray crystal; Yield: 65 %; mp 267–268 °C; IR (KBr) ν_{max} 3234, 3039, 2987, 1765, 1540, 1533, 1145 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz):$\delta = 2.47$ (s, 3H, $–CH_3$), 4.67

(s, 1H, –OH), 5.54 (s, 1H, –CH pyrimidine ring), 5.98 (s, 1H, –NH–C–Ph), 7.01–7.39 (m, 8H, Ar–H), 7.53 (s, 1H, –NH–N=CH), 7.57 (s, 1H, –NH–N=CH–), 8.43 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz):δ = 17.4 (–CH_3), 52.4 (–CH pyrimidine), 99.7 (–C=C–CH_3 pyrimidine ring), 115.5 (2), 116.9 (–C=C–F in phenyl ring), 125.2 (Ar–C), 127.4 (Ar–C), 129.5 (Ar–C) (2), 130.3 (Ar–C), 130.8 (Ar–C), 131.6 (Ar–C), 150.2 (–NHN=CH–), 155.3 (–CO pyrimidine ring), 155.9 (–C–CH_3), 158.5 (–C–OH), 161.6 (–C–F), 163.4 (–CONH–); LCMS *m/z*368 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16, H, 4.40, N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

4-(2-fluorophenyl)-N′-(4-hydroxybenzylidene)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (3e)

Light brown crystal; Yield: 68 %; mp 232–233 °C; IR (KBr) ν_{max} 3237, 3067, 2984, 1746, 1567, 1519, 1129 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz): δ = 2.49 (s, 3H, –CH_3), 5.23 (s, 1H, –OH), 5.59 (s, 1H, –CH pyrimidine ring), 6.04 (s, 1H, –NH–C–Ph), 7.11–7.47 (m, 8H, Ar–H), 7.68 (s, 1H, –NH–N=CH), 7.74 (s, 1H, –NH–N=CH–), 8.32 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 16.6 (–CH_3), 52.7 (–CH pyrimidine), 100.3 (–C=C–CH_3 pyrimidine ring), 115.3 (2), 116.5 (–C=C–F in phenyl ring), 125.5 (Ar–C), 127.7 (Ar–C), 129.9 (Ar–C) (2), 130.6 (Ar–C), 131.2 (Ar–C), 131.8 (Ar–C), 149.4 (–NHN=CH–), 155.8 (–CO pyrimidine ring), 156.5 (–C–CH_3), 158.7 (–C–OH), 161.4 (–C–F), 163.9 (–CONH–); LCMS *m/z*368 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

N′-(4-chlorobenzylidene)-4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (3f)

Light yellow crystal; Yield: 59 %; mp 276–277 °C; IR (KBr) ν_{max} 3278, 3043, 2976, 1763, 1549, 1533, 1131 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz): δ = 2.36 (s, 3H, –CH_3), 5.65 (s, 1H, –CH pyrimidine ring), 5.95 (s, 1H, –NH–C–Ph), 7.01–7.51 (m, 8H, Ar–H), 7.57 (s, 1H, –NH–C–Me), 7.72 (s, 1H, –NH–N=CH), 7.91 (s, 1H, –NH–N=CH–); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 17.4 (–CH_3), 52.7 (–CH pyrimidine), 100.3 (–C=C–CH_3 pyrimidine ring), 117.4 (–C=C–F in phenyl ring), 124.7(Ar–C), 129.5(Ar–C)(2), 130.4 (Ar–C) (2), 130.7 (Ar–C), 131.5 (Ar–C), 131.8 (Ar–C), 132.4 (Ar–C), 134.5 (Ar–C), 135.6 (–C–Cl), 149.7 (–NHN=CH–), 155.5 (–CO pyrimidine ring), 156.2 (–C–CH_3), 161.7 (–C–F), 163.7 (–CONH–); LCMS *m/z* 386 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

4-(2-fluorophenyl)-6-methyl-N′-(4-nitrobenzylidene)-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (3g)

Light orange crystal; Yield: 63 %; mp 287–288 °C; IR (KBr) ν_{max} 3265, 3042, 2967, 1751, 1537, 1523, 1129 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz): δ = 2.47 (s, 3H, –CH_3), 5.74 (s, 1H, –CH pyrimidine ring), 5.98 (s, 1H, –NH–C–Ph), 6.44 (s, 1H, –NH–N=CH), 7.04–7.57 (m, 8H, Ar–H), 7.59 (s, 1H, –NH–C–Me), 7.85 (s, 1H, –NH–N=CH–); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 16.8 (–CH_3), 51.8 (–CH pyrimidine), 99.9 (–C=C–CH_3 pyrimidine ring), 116.7 (–C=C–F in phenyl ring), 124.4 (Ar–C) (2), 126.1 (Ar–C), 128.3 (Ar–C) (2), 130.4 (Ar–C), 130.8 (Ar–C), 131.3 (Ar–C), 140.4, 147.7 (–C–NO_2), 149.4 (–NHN=CH–), 155.3 (–CO pyrimidine ring), 155.8 (–C–CH_3), 161.4 (–C–F), 163.3 (–CONH–); LCMS *m/z*397 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

4-(2-fluorophenyl)-6-methyl-N′-(2-nitrobenzylidene)-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (3h)

Light gray crystal; Yield: 76 %; mp 194–195 °C; IR (KBr) ν_{max} 3267, 3055, 2967, 1770, 1557, 1547, 1130 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz): δ = 2.38 (s, 3H, –CH_3), 5.69 (s, 1H, –CH pyrimidine ring), 6.04 (s, 1H, –NH–C–Ph), 6.49 (s, 1H, –NH–N=CH), 7.13–7.67 (m, 8H, Ar–H), 7.72 (s, 1H, –NH–C–Me), 7.92 (s, 1H, –NH–N=CH–); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 16.4 (–CH_3), 52.4 (–CH pyrimidine), 102.2 (–C=C–CH_3 pyrimidine ring), 116.4 (–C=C–F in phenyl ring), 124.6 (Ar–C) (2), 127.4 (Ar–C), 128.7 (Ar–C) (2), 130.5 (Ar–C), 130.9 (Ar–C), 131.6 (Ar–C), 140.2 (Ar–C), 147.9 (–C–NO_2), 149.7 (–NHN=CH–), 155.5 (–CO pyrimidine ring), 156.2 (–C–CH_3), 161.7 (–C–F), 163.5 (–CONH–); LCMS *m/z* 397 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

4-(2-fluorophenyl)-6-methyl-N′-(3-nitrobenzylidene)-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (3i)

Light brown crystal; Yield: 81 %; mp 222–223 °C; IR (KBr) ν_{max} 3242, 3039, 2969, 1767, 1548, 1539, 1141 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz): δ = 2.42 (s, 3H, –CH_3), 5.49 (s, 1H, –CH pyrimidine ring), 5.78 (s, 1H, –NH–C–Ph), 6.34 (s, 1H, –NH–N=CH), 7.03–7.58 (m, 8H, Ar–H), 7.81 (s, 1H, –NH–C–Me), 7.99 (s, 1H, –NH–N=CH–); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 17.4 (–CH_3), 51.3 (–CH pyrimidine), 102.4 (–C=C–CH_3 pyrimidine ring), 115.9 (–C=C–F in phenyl ring), 125.7 (Ar–C) (2), 127.9 (Ar–C), 128.8 (Ar–C) (2), 130.8 (Ar–C), 131.4 (Ar–C), 131.9 (Ar–C), 140.7 (Ar–C), 147.5 (–C–NO_2),

150.3 (–NHN=CH–), 155.4 (–CO pyrimidine ring), 155.9 (–C–CH_3), 161.5 (–C–F), 163.9 (–CONH–); LCMS *m/z* 397 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

4-(2-fluorophenyl)-N′-(4-methoxybenzylidene)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (3j)

Light pink crystal; Yield: 70 %; mp 237–238 °C; IR (KBr) ν_{max} 3252, 3057, 2979, 1767, 1549, 1531, 1145 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 2.53 (s, 3H, –CH_3), 3.78 (s, 3H, –OCH_3), 5.47 (s, 1H, –CH pyrimidine ring), 5.73 (s, 1H, –NH–C–Ph), 6.98–7.45 (m, 8H, Ar–H), 7.52 (s, 1H, –NH–C–Me), 7.67 (s, 1H, –NH–N=CH), 8.32 (s, 1H, –NH–N=CH–); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 16.9 (–CH_3), 51.5 (–CH pyrimidine), 57.8 (–OCH_3), 100.4 (–C=C–CH_3 pyrimidine ring), 114.7 (Ar–C) (2), 117.3 (–C=C–F in phenyl ring), 125.2 (Ar–C), 127.4 (Ar–C), 129.4 (Ar–C) (2), 130.4 (Ar–C), 130.7 (Ar–C), 131.4 (Ar–C), 149.7 (–NHN=CH–), 155.7 (–CO pyrimidine ring), 156.5 (–C–CH_3), 159.7 (–C–OCH_3), 161.8 (–C–F), 164.4 (–CONH–); LCMS *m/z* 382 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C,61.16; H,4.40; N,13.59; Found: C, 63.67; H, 4.83; N, 14.65.

4-(2-fluorophenyl)-N′-(2-methoxybenzylidene)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (3k)

Yellowish orange crystal; Yield: 60 %; mp 262–263 °C; IR (KBr) ν_{max} 3259, 3059, 2981, 1749, 1554, 1534, 1143 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 2.48 (s, 3H, –CH_3), 3.57 (s, 3H, –OCH_3), 5.54 (s, 1H, –CH pyrimidine ring), 5.81 (s, 1H, –NH–C–Ph), 7.03–7.38 (m, 8H, Ar–H), 7.59 (s, 1H, –NH–C–Me), 7.74 (s, 1H, –NH–N=CH), 8.24 (s, 1H, –NH–N=CH–); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 17.4 (–CH_3), 52.4 (–CH pyrimidine), 58.5 (–OCH_3), 100.7 (–C=C–CH_3 pyrimidine ring), 115.6 (Ar–C) (2), 116.7 (–C=C–F in phenyl ring), 125.8 (Ar–C), 128.5 (Ar–C), 129.7 (Ar–C) (2), 130.6 (Ar–C), 130.9 (Ar–C), 131.2 (Ar–C), 149.5 (–NHN=CH–), 155.5 (–CO pyrimidine ring), 156.3 (–C–CH_3), 160.3 (–C–OCH_3), 161.4 (–C–F), 163.7 (–CONH–); LCMS *m/z* 382 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

N′-(2,6-dichlorobenzylidene)-4-(2-fluorophenyl)-6-methyl-2-oxo-1,2,3,4-tetrahydropyrimidine-5-carbohydrazide (3l)

Light gray crystal; Yield: 79 %; mp 292–293 °C; IR (KBr) ν_{max} 3267, 3057, 2990, 1776, 1560, 1545, 1156 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 2.34 (s, 3H, –CH_3), 5.34 (s, 1H, –CH pyrimidine ring), 6.05 (s, 1H, –NH–C–Ph), 6.84–7.34 (m, 7H, Ar–H), 7.64 (s, 1H, –NH–C–Me), 8.45 (s, 1H, –NH–N=CH), 8.24 (s, 1H, –NH–N=CH–); ^{13}C NMR (DMSO-d_6, 100 MHz):δ = 16.6 (–CH_3), 51.7 (–CH pyrimidine), 99.5 (–C=C–CH_3 pyrimidine ring), 116.4 (–C=C–F in phenyl ring), 125.0 (Ar–C), 128.7 (Ar–C), 129.7 (Ar–C) (2), 130.4 (Ar–C), 130.7 (Ar–C), 131.4 (Ar–C), 132.4 (Ar–C), 133.7 (2) (–C–Cl), 140.5 (–NHN=CH–), 155.7 (–CO pyrimidine ring), 155.9 (–C–CH_3), 161.7 (–C–F), 163.9 (–CONH–); LCMS *m/z* 421 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

General procedure for the preparation of 5-(4-acetyl-5-aryl-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-ones (4a–l)

Acetic anhydride (0.02 mol) was added to compounds **(3a-l)** (0.01 mol), and the reaction mass was refluxed for 2 h using reflux condenser. After completion of reaction, the reaction mixture was poured into ice-cold water. The precipitate was filtered off, washed with water, dried and recrystallized from DMF–ethanol to give product **(4a-l)**.

5-(4-acetyl-5-phenyl-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-one (4a)

Light pink crystal; Yield: 72 %; mp 240–241 °C; IR (KBr) ν_{max} 3210, 3095, 2924, 1751, 1594,1557, 1140 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 2.09 (s, 3H, –$COCH_3$), 2.45 (s, 3H, –CH_3), 5.56 (s, 1H, –CH pyrimidine ring), 5.93 (s, 1H, –NH–C–Ph), 6.98 (s, 1H, –CH oxadiazole ring), 7.09–7.39 (m, 9H, Ar–H), 7.91 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 16.3 (–CH_3), 23.2 (–$COCH_3$), 55.3 (–CH pyrimidine), 93.5 (–C=C–CH_3 pyrimidine ring), 101.0 (–CH of oxadiazole ring), 116.7 (–C=C–F in phenyl ring), 127.5 (Ar–C) (2), 128.4 (Ar–C) (2), 128.7 (Ar–C), 128.9 (Ar–C) (2), 130.2 (Ar–C), 134.3 (Ar–C), 140.7, 155.5 (–CO pyrimidine ring), 156.2 (–C–CH_3), 158.2 (–$COCH_3$), 160.9 (–C=N oxadiazole ring), 161.5 (–C–F); LCMS *m/z* 394 $[M]^+$; Anal. calcd. for $C_{21}H_{19}FN_4O_3$: C, 63.95; H, 4.86; N, 14.21; Found: C, 63.07; H, 4.83; N, 14.65.

5-(4-acetyl-5-(2-fluorophenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-one (4b)

Light brown crystal; Yield: 58 %; mp 254–255 °C; IR (KBr) ν_{max} 3220, 3087, 2916, 1745, 1578,1567, 1149 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 2.15 (s, 3H, –$COCH_3$), 2.45 (s, 3H, –CH_3), 5.56 (s, 1H, –CH pyrimidine ring), 5.89 (s, 1H, –NH–C–Ph), 6.98 (s, 1H, –CH oxadiazole ring), 6.99–7.41 (m, 8H, Ar–H), 7.85 (s,

1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 16.7 (–CH_3), 24.3 (–$COCH_3$), 53.5 (–CH pyrimidine), 95.7 (–C=C–CH_3 pyrimidine ring), 97.0 (–CH of oxadiazole ring), 116.9 (–C=C–F in phenyl ring), 117.5 (Ar–C), 124.5 (Ar–C), 124.8 (Ar–C), 125.3 (Ar–C), 128.7(Ar–C), 129.9 (Ar–C), 130.7 (Ar–C), 131.3 (Ar–C), 131.5 (Ar–C), 155.8 (–CO pyrimidine ring), 156.7 (–C–CH_3), 158.5 (–$COCH_3$), 161.3 (–C=N oxadiazole ring), 161.5, 161.8 (–C–F); LCMS *m/z* 412 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 61.14; H, 4.70; N, 13.56.

5-(4-acetyl-5-(4-fluorophenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-one (4c)

Light gray crystal; Yield: 69 %; mp 160–161 °C; IR (KBr) ν_{max} 3235, 3078, 2934, 1745(–CO stret.), 1580,1561, 1136 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz): δ = 2.26 (s, 3H, –$COCH_3$), 2.34 (s, 3H, –CH_3), 5.39 (s, 1H, –CH pyrimidine ring), 5.76 (s, 1H, –NH–C–Ph), 6.87 (s, 1H, –CH oxadiazole ring), 7.10–7.49 (m, 8H, Ar–H), 7.92 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 16.9 (–CH_3), 24.4 (–$COCH_3$), 53.8 (–CH pyrimidine), 94.5 (–C=C–CH_3 pyrimidine ring), 98.8 (–CH of oxadiazole ring), 118.8 (–C=C–F in phenyl ring), 120.8 (Ar–C), 123.8 (Ar–C), 124.4 (Ar–C), 125.7 (Ar–C), 127.5 (Ar–C), 128.4 (Ar–C), 129.4 (Ar–C), 130.3 (Ar–C), 130.5 (Ar–C), 155.6 (–CO pyrimidine ring), 156.8 (–C–CH_3), 158.4 (–$COCH_3$), 161.2 (–C=N oxadiazole ring), 161.5, 161.7 (–C–F); LCMS *m/z* 412 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 61.17; H, 4.27; N, 13.68.

5-(4-acetyl-5-(2-hydroxyphenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-one (4d)

Light orange crystal; Yield: 56 %; mp 189–190 °C; IR (KBr) ν_{max} 3445, 3226, 3067, 2945, 1739, 1584, 1556, 1158 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz): δ = 2.35 (s, 3H, –$COCH_3$), 2.46 (s, 3H, –CH_3), 5.43 (s, 1H, –CH pyrimidine ring), 5.67 (s, 1H, –NH–C–Ph), 6.94 (s, 1H, –CH oxadiazole ring), 7.04–7.39 (m, 8H, Ar–H), 7.84 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 16.4 (–CH_3), 23.4 (–$COCH_3$), 53.6 (–CH pyrimidine), 94.7 (–C=C–CH_3 pyrimidine ring), 97.3 (–CH of oxadiazole ring), 117.5 (–C=C–F in phenyl ring), 120.4 (Ar–C), 123.4 (Ar–C), 124.6 (Ar–C), 126.3 (Ar–C), 127.7 (Ar–C), 128.7 (Ar–C), 129.5 (Ar–C), 130.6 (Ar–C), 131.7 (Ar–C), 154.6 (–CO pyrimidine ring), 156.4 (–C–CH_3), 158.7 (–$COCH_3$), 161.3 (–C=N oxadiazole ring), 161.2, 162.3 (–C–F); LCMS *m/z* 410$[M]^+$; Anal. calcd. for $C_{21}H_{19}FN_4O_4$: C, 61.46; H, 4.67; N, 13.65; Found: C, 61.48; H, 4.51; N, 13.19.

5-(4-acetyl-5-(4-hydroxyphenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-one (4e)

Light yellow crystal; Yield: 75 %; mp 286–287 °C; IR (KBr) ν_{max} 3456, 3245, 3078, 2950, 1729, 1578, 1539, 1178 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz): δ = 2.29 (s, 3H, –$COCH_3$), 2.39 (s, 3H, –CH_3), 5.56 (s, 1H, –CH pyrimidine ring), 5.78 (s, 1H, –NH–C–Ph), 6.99 (s, 1H, –CH oxadiazole ring), 7.08–7.45 (m, 8H, Ar–H), 8.04 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 18.2 (–CH_3), 24.6 (–$COCH_3$), 53.8 (–CH pyrimidine), 94.5 (–C=C–CH_3 pyrimidine ring), 96.5 (–CH of oxadiazole ring), 116.4 (–C=C–F in phenyl ring), 120.6 (Ar–C), 123.7 (Ar–C), 124.3 (Ar–C), 126.5 (Ar–C), 127.9 (Ar–C), 128.5 (Ar–C), 129.8 (Ar–C), 130.4 (Ar–C), 131.9 (Ar–C), 154.8 (–CO pyrimidine ring), 156.7 (–C–CH_3), 158.4 (–$COCH_3$), 161.6 (–C=N oxadiazole ring), 161.8, 162.5 (–C–F); LCMS *m/z* 410 $[M]^+$; Anal. calcd. for $C_{21}H_{19}FN_4O_4$: C, 61.46, H, 4.67, N, 13.65; Found: C, 61.62, H, 4.84, N, 13.16.

5-(4-acetyl-5-(4-chlorophenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-one (4f)

Light yellow crystal; Yield: 57 %; mp 257–258 °C; IR (KBr) ν_{max} 3252, 3082, 2962, 1734, 1584, 1543, 1157 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz): δ = 2.32 (s, 3H, –$COCH_3$), 2.43 (s, 3H, –CH_3), 5.62 (s, 1H, –CH pyrimidine ring), 5.82 (s, 1H, –NH–C–Ph), 6.87 (s, 1H, –CH oxadiazole ring), 7.12–7.39 (m, 8H, Ar–H), 7.83 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 16.4 (–CH_3), 23.5 (–$COCH_3$), 54.4 (–CH pyrimidine), 93.7 (–C=C–CH_3 pyrimidine ring), 95.7 (–CH of oxadiazole ring), 116.5 (–C=C–F in phenyl ring), 120.3 (Ar–C), 123.6 (Ar–C), 124.8 (Ar–C), 126.4 (Ar–C), 127.7 (Ar–C), 128.4 (Ar–C), 129.3 (Ar–C), 130.7 (Ar–C), 131.6 (Ar–C), 154.5 (–CO pyrimidine ring), 156.4 (–C–CH_3), 158.7 (–$COCH_3$), 161.3 (–C=N oxadiazole ring), 161.4, 162.8 (–C–F); LCMS *m/z* 428 $[M]^+$; Anal. calcd. for $C_{21}H_{18}FClN_4O_3$: C, 58.82; H, 4.23; N, 13.06; Found: C, 58.31; H, 4.57; N, 13.76.

5-(4-acetyl-5-(2-nitrophenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-one (4g)

Light yellow crystal; Yield: 59 %; mp 234–235 °C; IR (KBr) ν_{max} 3247, 3072, 2968, 1743, 1538, 1551,

1162 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 2.23 (s, 3H, –$COCH_3$), 2.42 (s, 3H, –CH_3), 5.53 (s, 1H, –CH pyrimidine ring), 6.04 (s, 1H, –NH–C–Ph), 6.91 (s, 1H, –CH oxadiazole ring), 7.20–8.10 (m, 8H, Ar–H), 8.15 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 16.7 (–CH_3), 23.4 (–$COCH_3$), 55.7 (–CH pyrimidine), 94.4 (–C=C–CH_3 pyrimidine ring), 96.3 (–CH of oxadiazole ring), 115.7 (–C=C–F in phenyl ring), 124.6 (Ar–C) (2), 125.2 (Ar–C), 126.7 (Ar–C) (2), 130.5 (Ar–C), 131.2 (Ar–C), 131.5 (Ar–C), 141.9, 148.5 (C–NO_2), 155.8 (–CO pyrimidine ring), 156.5 (–C–CH_3), 158.9 (–$COCH_3$), 161.3 (–C=N oxadiazole ring), 162.8 (–C–F); LCMS *m/z* 439 $[M]^+$; Anal. calcd. for $C_{21}H_{18}FN_5O_5$: C, 57.40; H, 4.13; N, 15.94; Found: C, 57.37; H, 4.23; N, 15.91.

5-(4-acetyl-5-(3-nitrophenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-one (4h)

Light yellow crystal; Yield: 59 %; mp 206–207 °C; IR (KBr) ν_{max} 3254, 3067, 2974, 1756, 1544, 1557, 1155 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 2.17 (s, 3H, –$COCH_3$), 2.35 (s, 3H, –CH_3), 5.49 (s, 1H, –CH pyrimidine ring), 6.14 (s, 1H, –NH–C–Ph), 6.88 (s, 1H, –CH oxadiazole ring), 7.14–7.89 (m, 8H, Ar–H), 8.04 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 17.4 (–CH_3), 24.4 (–$COCH_3$), 55.5 (–CH pyrimidine), 94.7 (–C=C–CH_3 pyrimidine ring), 96.7 (–CH of oxadiazole ring), 116.6 (–C=C–F in phenyl ring), 124.9 (Ar–C) (2), 125.5 (Ar–C), 126.4 (Ar–C) (2), 130.7 (Ar–C), 131.4 (Ar–C), 131.8 (Ar–C), 142.4, 148.7 (C–NO_2), 155.4 (–CO pyrimidine ring), 156.7 (–C–CH_3), 158.4 (–$COCH_3$), 161.7 (–C=N oxadiazole ring), 162.4 (–C–F); LCMS *m/z* 439 $[M]^+$; Anal. calcd. for $C_{21}H_{18}FN_5O_5$: C, 57.40; H, 4.13; N, 15.94; Found: C, 57.69; H, 4.68; N, 15.56.

5-(4-acetyl-5-(4-nitrophenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-one (4i)

Light yellow crystal; Yield: 66 %; mp 222–223 °C; IR (KBr) ν_{max} 3247, 3072, 2956), 1767, 1546, 1534, 1167 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 2.17 (s, 3H, –$COCH_3$), 2.39 (s, 3H, –CH_3), 5.55 (s, 1H, –CH pyrimidine ring), 6.18 (s, 1H, –NH–C–Ph), 6.93 (s, 1H, –CH oxadiazole ring), 7.18–7.74 (m, 8H, Ar–H), 8.13 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 17.6 (–CH_3), 23.3 (–$COCH_3$), 54.9 (–CH pyrimidine), 94.3 (–C=C–CH_3 pyrimidine ring), 95.8 (–CH of oxadiazole ring), 115.9 (–C=C–F in phenyl ring), 124.4 (Ar–C) (2), 125.7 (Ar–C), 126.7 (Ar–C) (2), 129.3 (Ar–C), 130.7 (Ar–C), 131.5 (Ar–C), 142.6, 147.6 (C–NO_2), 155.7 (–CO pyrimidine ring), 156.9 (–C–CH_3), 158.6 (–$COCH_3$), 160.3 (–C=N oxadiazole ring), 162.7 (–C–F); LCMS *m/z* 439 $[M]^+$; Anal. calcd. for $C_{21}H_{18}FN_5O_5$: C, 57.40; H,4.13; N,15.94; Found: C, 57.70; H, 4.77; N, 15.60.

5-(4-acetyl-5-(2,6-dichlorophenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-one (4j)

Light brown crystal; Yield: 78 %; mp 198–199 °C; IR (KBr) ν_{max} 3246, 3058, 2979, 1729, 1557, 1537, 1148 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 2.14 (s, 3H, –$COCH_3$), 2.54 (s, 3H, –CH_3), 5.59 (s, 1H, –CH pyrimidine ring), 6.05 (s, 1H, –NH–C–Ph), 6.88 (s, 1H, –CH oxadiazole ring), 7.11–7.55 (m, 7H, Ar–H), 7.70 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 17.4 (–CH_3), 23.4 (–$COCH_3$), 54.6 (–CH pyrimidine), 94.5 (–C=C–CH_3 pyrimidine ring), 95.6 (–CH of oxadiazol ring), 116.7 (–C=C–F in phenyl ring), 124.8 (Ar–C), 129.7 (Ar–C) (2), 130.3 (Ar–C), 130.8 (Ar–C) (2), 131.5 (Ar–C), 132.6 (Ar–C), 134.5 (Ar–C) (2), 156.5 (–CO pyrimidine ring), 157.7 (–C–CH_3), 158.2 (–$COCH_3$), 160.6 (–C=N oxadiazole ring), 161.3 (–C–F); LCMS *m/z* 462 $[M]^+$; Anal. calcd. for $C_{21}H_{17}Cl_2FN_4O_3$: C, 54.44; H, 3.70; N, 12.09; Found: C, 54.71; H, 3.79; N, 12.01.

5-(4-acetyl-5-(4-methoxyphenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-one (4k)

Light brown crystal; Yield: 72 %; mp 256–257 °C; IR (KBr) ν_{max} 3267, 3047, 2963, 1761, 1548, 1535, 1153 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 2.03 (s, 3H, –$COCH_3$), 2.44 (s, 3H, –CH_3), 3.78 (s, 3H, –OCH_3), 5.58 (s, 1H, –CH pyrimidine ring), 6.09 (s, 1H, –NH–C–Ph), 6.97 (s, 1H, –CH oxadiazole ring), 6.99–7.39 (m, 8H, Ar–H), 7.74 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 16.8 (–CH_3), 24.7 (–$COCH_3$), 53.7 (–CH pyrimidine), 56.3 (–OCH_3), 93.9 (–C=C–CH_3 pyrimidine ring), 101.0 (–CH of oxadiazole ring), 114.0 (2), 116.4 (–C=C–F in phenyl ring), 125.0 (Ar–C), 127.1 (Ar–C) (2), 129.7(Ar–C), 130.6 (Ar–C), 131.3 (Ar–C), 131.6 (Ar–C), 156.3 (–CO pyrimidine ring), 156.7 (–C–CH_3), 158.3 (–$COCH_3$), 161.2 (–C=N oxadiazole ring), 161.7 (–C–F), 162.7 (–C–OCH_3); LCMS *m/z* 424 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

5-(4-acetyl-5-(2-methoxyphenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-4-(2-fluorophenyl)-6-methyl-3,4-dihydropyrimidin-2(1H)-one (4l)

Light brown crystal; Yield: 72 %; mp 240–241 °C; IR (KBr) ν_{max} 3254, 3052, 2975, 1759, 1544, 1529,

1137 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz): δ = 2.09 (s, 3H, –$COCH_3$), 2.47 (s, 3H, –CH_3), 3.67 (s, 3H, –OCH_3), 5.63 (s, 1H, –CH pyrimidine ring), 6.13 (s, 1H, –NH–C–Ph), 6.95 (s, 1H, –CH oxadiazole ring), 7.02–7.43 (m, 8H, Ar–H), 7.76 (s, 1H, –NH–C–Me); ^{13}C NMR (DMSO-d_6, 100 MHz): δ = 16.8 (–CH_3), 24.7(–$COCH_3$), 53.7 (–CH pyrimidine), 56.3 (–OCH_3), 93.9 (–C=C–CH_3 pyrimidine ring), 101.0 (–CH of oxadiazole ring), 114.0 (2), 116.4 (–C=C–F in phenyl ring), 125.0 (Ar–C), 127.1 (Ar–C) (2), 129.7 (Ar–C), 130.6 (Ar–C), 131.3 (Ar–C), 131.6 (Ar–C), 156.3 (–CO pyrimidine ring), 156.7 (–C–CH_3), 158.3 (–$COCH_3$), 161.2 (–C=N oxadiazole ring), 161.7 (–C–F), 162.7 (–C–OCH_3); LCMS *m/z* 424 $[M]^+$; Anal. calcd. for $C_{21}H_{18}F_2N_4O_3$: C, 61.16; H, 4.40; N, 13.59; Found: C, 63.67; H, 4.83; N, 14.65.

Biological assay

Antitubercular assay

Antitubercular activity determination of the test compounds against *M. tuberculosis* $H_{37}Rv$ was performed by slightly modified Lowenstein–Jensen method described earlier (Kathrotiya and Patel, 2013), where 250 μg/mL dilution of each test compound prepared in DMSO was added in liquid Lowenstein–Jensen medium, and then media was sterilized by inspissation method. A culture of *M. tuberculosis* $H_{37}Rv$ growing on Lowenstein–Jensen medium was harvested in 0.85 % saline in bijou bottles. These tubes were then incubated at 37 °C for 24 h followed by streaking of *M. tuberculosis* $H_{37}Rv$ (5×10^4 bacilli per tube). These tubes were then incubated at 37 °C. Growth of bacilli was seen after 12, 22 days and finally 28 days of incubation. Tubes having the compounds were compared with control tubes where medium alone was incubated with *M. tuberculosis* $H_{37}Rv$. The concentration at which no development of colonies occurred or <20 colonies was taken as MIC concentration of test compound. The screening results are summarized in Table 1 as % inhibition and MIC values relative to standard drug isoniazid.

Conclusion

In conclusion, we have conveniently synthesized a novel series of 1,3,4-oxadiazole clubbed dihydropyrimidines in very good yields and evaluated them for in vitro antitubercular activity with anticipation of generating new structural leads serving as potential antitubercular agents. On the basis of structure activity relationship studies, it was observed that compounds **4c**, **4f** and **4i** with electronic withdrawing group/atoms such as nitro, chloro and fluoro at *para* position of 1,3,4-oxadiazolyl phenyl ring showed very good antitubercular activity. Compound **4i** substituted with fluoro group emerged as the most potent antitubercular agents with MIC equivalent to isoniazid. Consequently, such type of compounds would represent a fertile matrix for further development of novel and potent antitubercular agents requiring further optimization in order to discover the scope and limitation of its biological profile.

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