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Synthesis and biological evaluation of novel 6-substituted 5-alkyl-2-(arylcarbonylmethylthio)pyrimidin-4(3*H*)-ones as potent non-nucleoside HIV-1 reverse transcriptase inhibitors

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Abstract—A novel series of 2-arylcarbonylmethylthio-6-arylmethylpyrimidin-4(3*H*)-ones have been synthesized and evaluated for in vitro anti-HIV activities in MT-4 cells. Most of these new compounds showed moderate to potent activities against wild-type HIV-1 with an EC_{50} range from 8.97 μM to 0.010 μM. Among them, the 6-(3,5-dimethylbenzyl) analogue **5p** was identified as the most promising compound (EC_{50} = 0.010 μM, SI > 31,800) associated with moderate activity against the HIV-1 double mutant RT strain K103N + Y181C. The structure–activity relationships of these new congeners were further discussed.

1. Introduction

Over the past decade, 2-alkylthio-6-benzylpyrimidin-4(3*H*)-ones (*S*-DABOs)[1] have been the subject of great interest and have led in recent years to the identification of several new structures (**2–4**, Fig. 1)[2–11] displaying excellent activities as non-nucleoside inhibitors of HIV-1 reverse transcriptase (RT). Our recent work on *S*-DABOs disclosed that the assembly of an arylcarbonylmethyl moiety at the C-2 sulfur atom bound to the pyrimidine nucleus has resulted in the synthesis of many new analogues (**4**, Fig. 1)[8] with potent antiviral activities against HIV-1. These results indicated that this subclass of *S*-DABO analogues raises the appealing possibility for further structural modifications using **4** as the starting point. In the course of our search for new anti-HIV-1 agents, we became interested in investigating the effect of the substituent at position 6 of the 2-arylcarbonylmethythio substituted *S*-DABOs (**5**, Fig. 1) on anti-HIV activity with the aim to delineate the structure–activity relationships (SAR) of *S*-DABOs and to further improve their HIV-1 inhibitory potency. Herein, the detailed synthesis, anti-HIV-1 activity and preliminary SAR studies of these new congeners are described.

2. Chemistry

The synthesis of the target compounds **5a–q** was straightforward and is depicted in Scheme 1. The substituted β-ketoesters **7a–p** were easily prepared using the

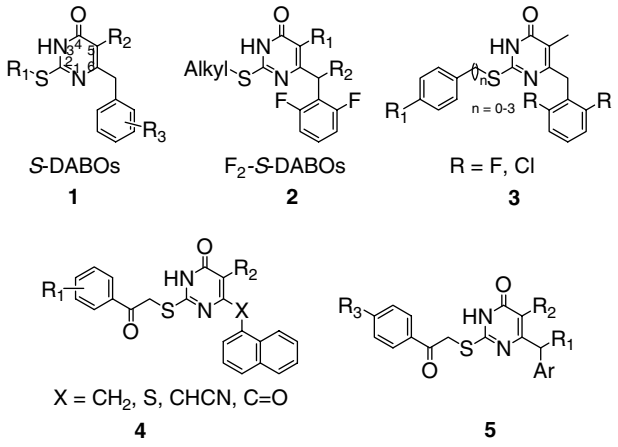

Figure 1. Chemical structures of *S*-DABOs.

Keywords: HIV; NNRTIs; SAR; *S*-DABOs.
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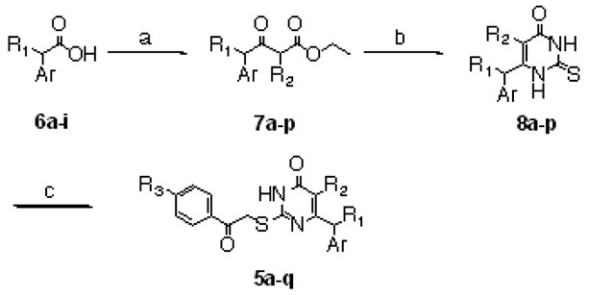

6,7,8	Ar	R_1	R_2
a	2-Br-Ph	H	Me
b	3-Br-Ph	H	Me
c	3-OH-Ph	H	Me
d	4-Br-Ph	H	Me
e	3,5-$(Me)_2$-Ph	H	Me
f	3,5-$(CF_3)_2$-Ph	H	Me
g	2,6-Cl_2-Ph	H	Me
h	Ph	Ph	Me
i	1-naphthyl	H	Me

7, 8	Ar	R_1	R_2
j	2-Br-Ph	H	Et
k	3-Br-Ph	H	Et
l	4-Br-Ph	H	Et
m	4-Ph-Ph	H	Et
n	3,5-$(Me)_2$-Ph	H	Et
o	3,5-$(CF_3)_2$-Ph	H	Et
p	Ph	Ph	Et

5	Ar	R_1	R_2	R_3
a	2-Br-Ph	H	Me	MeO
b	3-Br-Ph	H	Me	MeO
c	3-OH-Ph	H	Me	MeO
d	4-Br-Ph	H	Me	MeO
e	3,5-$(Me)_2$-Ph	H	Me	MeO
f	3,5-$(CF_3)_2$-Ph	H	Me	MeO
g	2,6-Cl_2-Ph	H	Me	MeO
h	Ph	Ph	Me	MeO
i	1-naphthyl	H	Me	H

5	Ar	R_1	R_2	R_3
j	2-Br-Ph	H	Et	MeO
k	3-Br-Ph	H	Et	MeO
l	4-Br-Ph	H	Et	MeO
m	4-Ph-Ph	H	Et	MeO
n	3,5-$(Me)_2$-Ph	H	Et	MeO
o	3,5-$(CF_3)_2$-Ph	H	Et	MeO
p	3,5-$(Me)_2$-Ph	H	Et	H
q	Ph	Ph	Et	MeO

Scheme 1. Synthesis of compounds **5a–q**. Reagents and conditions: (a) i—CDI, CH_3CN, rt, 30 min; ii— $R_2CH(CO_2Et)(CO_2K)$, Et_3N, anhydrous $MgCl_2$, rt, overnight, then reflux, 2 h. (b) Thiourea, EtONa, EtOH, reflux, 10 h. (c) R_3COCH_2X (X = Br or Cl), K_2CO_3, DMF, rt, 24 h.

method of Clay[12] by exposure of substituted arylacetic acids **6a–i** to 1,1′-carbonyldiimidazole (CDI) followed by treatment with different ethyl potassium malonate in the presence of anhydrous $MgCl_2$ and Et_3N, in which the requisite starting materials **6a–i** were commercially available or could be readily prepared. Subsequent condensation of **7a–p** with thiourea in the presence of EtONa in refluxing EtOH led to the key intermediates **8a–p** according to our previously reported procedure.[13] Next, selective S-alkylation of **8a–p** with the appropriate arylcarbonylmethyl halides (1:1.1) in the presence of K_2CO_3 in anhydrous DMF afforded the desirable target compounds **5a–q**. It is worth noting that in this reaction, some by-products such as *N*-substituted or *O*-substituted or di-substituted compounds were also formed. However, the *S*-alkylated compounds **5a–q** were the main products, which could be easily purified by column chromatography or by crystallization.

All the target compounds **5a–q** were characterized by NMR, MS, IR and elemental analysis. Both analytical and spectral data of all the compounds are in full agreement with the proposed structures. Moreover, comparison of the spectroscopic data of the new compounds with those of the previously reported analogues[8] further confirmed the above structures.

3. Results and discussion

All of the newly synthesized *S*-DABO analogues (**5a–q**) were first evaluated for their cytotoxicity and anti-HIV activity in MT-4 cells infected with wild-type HIV-1 strain III_B and HIV-2 strain ROD. The results, expressed as CC_{50}, EC_{50} and SI, are summarized in Table 1 together with those of efavirenz, delavirdine, AZT, and DDI as reference drugs.

As can be seen from Table 1, most of the test compounds inhibited HIV-1 replication in a lower micromolar concentration range. It is worth noting that a strikingly low cytotoxicity of these new *S*-DABOs was observed; and the majority of them were non-cytotoxic for MT-4 cells at doses as high as 236 μM. Among the highly active compounds, 6-(3,5-dimethylbenzyl)-5-ethyl-2-(phenylcarbonylmethylthio)pyrimidin-4(3*H*)-one (**5p**) turned out to be the most potent inhibitor with an EC_{50} of 0.010 μM against HIV-1 being about 7.2-times higher than that of delavirdine. More importantly, it also exhibited a high selectivity index (SI > 31,800). Some other compounds, **5e**, **5g**, **5j**, and **5n**, also showed high anti-HIV-1 potency (EC_{50} = 0.054, 0.044, 0.040, and 0.018 μM, respectively) and excellent selectivity indices (SI = 5704, 6318, 4675, and 13278, respectively).

Table 1. Biological activities of compounds **5a–q** against HIV-1 and HIV-2 in MT-4 cells[a]

Compound	Ar	R1	R2	R3	EC_{50}[b] (μM)		CC_{50}[c] (μM)	SI[d] (HIV-1 III_B)
					HIV-1 III_B	HIV-2 ROD		
5a	2-Br–Ph	H	Me	MeO	0.63 ± 0.57	>272	>272	>432
5b	3-Br–Ph	H	Me	MeO	0.89 ± 0.01	203	⩾196	⩾220
5c	3-OH–Ph	H	Me	MeO	4.44 ± 0.38	>59	70 ± 22	16
5d	4-Br–Ph	H	Me	MeO	8.97 ± 1.65	⩾226	>272	>30
5e	3,5-Me_2–Ph	H	Me	MeO	0.054 ± 0.008	60.62 ± 29.97	>308	>5704
5f	3,5-$(CF_3)_2$–Ph	H	Me	MeO	46.06 ± 39.64	>242	>242	>5
5g	2,6-Cl_2–Ph	H	Me	MeO	0.044 ± 0.006	>278	>278	>6318
5h	Ph	Ph	Me	MeO	1.38 ± 0.27	>274	>274	>199
5i	1-Naphthyl	H	Me	H	0.22 ± 0.04	⩾42.20	⩾305	⩾1386
5j	2-Br–Ph	H	Et	MeO	0.040 ± 0.001	>187	187 ± 19	4675
5k	3-Br–Ph	H	Et	MeO	0.16 ± 0.01	>142	142 ± 13	887
5l	4-Br–Ph	H	Et	MeO	7.94 ± 2.41	>264	>264	>33
5m	4-Ph–Ph	H	Et	MeO	>266	>266	>266	na[e]
5n	3,5-Me_2–Ph	H	Et	MeO	0.018 ± 0.013	52.78 ± 6.04	⩾239	⩾13,278
5o	3,5-$(CF_3)_2$–Ph	H	Et	MeO	0.79 ± 0.008	>236	>236	299
5p	3,5-Me_2–Ph	H	Et	H	0.010 ± 0.002	154 ± 135	>318	>31,800
5q	Ph	Ph	Et	MeO	3.49 ± 3.10	>266	>266	76
Efavirenz					0.003		>6.34	2113
Delavirdine					0.072		>3.62	>50
AZT					0.005		>93.55	>17,990
DDI					5.37 ± 0.1	2.71 ± 0.25	⩾529	⩾98

[a] All data represent mean values for at least two separate experiments.
[b] EC_{50}, effective concentration of compound required to protect the cell against viral cytopathogenicity by 50% in MT-4 cells.
[c] CC_{50}, cytotoxic concentration of compound that reduces the normal uninfected MT-4 cell viability by 50%.
[d] SI, selectivity index; ratio CC_{50}/EC_{50}.
[e] na, not active.

These promising results further highlighted that the phenylcarbonylmethythio moiety was an optimal substituent at the C-2 position of the pyrimidine ring, and that highly potent HIV-1 inhibitors could be obtained within *S*-DABOs sharing these common structural features. Also, the results shown in Table 1 revealed some important SAR information on the role of different substitutions in the aromatic ring of the C-6 benzyl moiety.

In the case of monosubstituted analogues (**5a–5d**, and **5j–5m**), a clear positional preference for substitution on the phenyl ring was observed by direct comparison: the inhibitory potency was higher for the *ortho*-substituted analogues (**5a** and **5j**) than for the *meta*-substituted ones (**5b** and **5k**) which, in turn, was more active than the *para*-substituted congeners (**5d** and **5l**). This conclusion is in agreement with the previous relative structure–activity relationship studies.[4] Furthermore, disubstitutions with two chlorine atoms at the *ortho*-position of the phenyl ring led to compound **5g** capable of inhibiting HIV-1 replication at nanomolar concentration, suggesting that this type of substitution pattern is highly favored with regard to the HIV-1 inhibitory activity. The above finding is also in agreement with the earlier SAR studies[4,9] concerning the 2,6-dichlorophenyl moiety of *S*-DABOs, which have been suggested to exert their favorable effect on anti-HIV-1 activity by enhancement of the putative charge-transfer interactions between the π-stacking aromatic rings of the inhibitor and Y188, and Y181 in RT.

Although the SAR discussed above appeared to be very similar to those found in other series of *S*-DABOs (i.e., **2** and **3**), there emerged some strikingly different SAR attributes on the substitution pattern of the phenyl ring when more effective nanomolar HIV-1 inhibitors **5e**, **5n** and **5p** with exceptionally high SI index were considered. Clearly, their high activity and selectivity toward HIV-1 could be attributed to the introduction of two *meta*-methyl groups into the phenyl ring, whereas in other series of *S*-DABOs this kind of structural variations had negligible effects on HIV-1 inhibitory activity.[3,5] The underlying factors responsible for the significantly improved antiviral potency of these compounds remain to be elucidated. One explanation may be that the additional *meta*-dimethyl substituents on the aromatic ring are in an appropriate spatial orientation to make extensive favorable van der Waals contacts with the highly conserved residue W229 located at the hydrophobic binding pocket of RT.

As for C-5 modifications it has been demonstrated that variation at this position plays an important role in determining the activity of *S*-DABOs.[8,14] Consistent with the SAR for our lead compounds 2-arylcarbonylmethyl-6-naphthylmethyl-substituted *S*-DABOs (**4**), a significant increase (1.1- to 58-fold) of anti-HIV-1 potency was found for all the newly synthesized analogues except **5h** and **5q** whose C-5 methyl group was replaced with an ethyl group. It should be pointed out that the above observations contrasted sharply with the conclusion drawn by Artico et al. based on 2-alkyl-substituted *S*-DABOs (**2**), in which the steric bulkiness of C-5 substituent is detrimental to HIV-1 inhibitory activity (H ⩾ Me > Et > *i*-Pr).[5]

Overall, the discrepancy of SAR at position 5, together with 3,5-dimethylsubstitution on the C-6 phenyl ring

Table 2. Activity of selected derivatives **5e**, **5n** and **5p** against (K103N+Y181C) HIV-1 mutant strain in MT-4 cells[a]

Compound	Ar	R1	R2	R3	EC_{50}[b] (μM)		CC_{50}[c] (μM)	SI[d] (RES056)
					RES056[f]	HIV-1 III_B		
5e	3,5-Me_2–Ph	H	Me	OMe	>308	0.054 ± 0.008	>308	na[e]
5n	3,5-Me_2–Ph	H	Et	OMe	>239	0.018 ± 0.013	⩾239	na[e]
5p	3,5-Me_2–Ph	H	Et	H	19.82	0.010 ± 0.002	>318	>16
Efavirenz					0.38	0.003	>6.34	17
Delavirdine					>3.62	0.072	>3.62	na[e]
AZT					0.006	0.005	>93.55	>15,592

[a] All data represent mean values for at least two separate experiments.
[b–e] See legend to Table 1.
[f] RES056, HIV-1 mutated strain bearing both K103N andY181C mutations.

being the most favorable structural variation, further corroborates the rationale behind this study: 2-arylcarbonylmethyl-substituted *S*-DABOs might interact with RT in a different way compared to closely related analogues (e.g., **2**); and more potent HIV-1 inhibitors with improved antiviral profile, especially against HIV-1 resistant mutants, might be achieved by appropriate chemical modifications on these unique *S*-DABOs.[15]

In the light of these, we further evaluated the capability of compounds **5e**, **5n** and **5p** to inhibit the replication of HIV-1 strain (RES056) bearing both NNRTI-characteristic K103N and Y181C mutations in cell culture (Table 2). It was found that compound **5p** exhibited some activity against the mutant strain at the lower micromolar concentrations.

In addition, all the title compounds were also tested for their antiviral activity against HIV-2 in MT-4 cells, but none was found effective except for compounds **5e**, **5i**, and **5n** displaying moderate potency at micromolar concentrations that were close to their toxicity/solubility threshold (Table 1).

4. Conclusions

In summary, a good agreement between our newly synthesized analogues and other series of *S*-DABOs was observed when the phenyl ring of the C-6 benzyl moiety was substituted with halogens. In particular, compound **5g** containing two chlorine atoms at the *ortho*-position exhibited potent anti-HIV-1 activity (EC_{50} = 0.044 μM, SI > 6318). However, the most favorable structural variation in this new series of *S*-DABOs turned out to be the introduction of two methyl groups into the *meta*-position of the phenyl ring leading to more potent and selective HIV-1 inhibitors **5e**, **5n** and **5p** (EC_{50} = 0.054, 0.018, and 0.010 μM, SI > 5704, 13278, and 31,800, respectively), while such variations had little effects in other series of *S*-DABOs. These peculiar and promising results imply that this new series of *S*-DABOs may interact with RT in a different way compared to the closely related analogues (**2**). Thus, 2-arylcarbonylmethylthio-6-(3,5-dimethylbenzyl)substituted-*S*-DABOs, especially compound **5p**, show great potential as candidates for further development of new HIV-1 specific inhibitors with an improved antiviral profile.

5. Experimental

5.1. Chemistry

5.1.1. General. Melting points were measured on a WRS-1 digital melting point apparatus and are uncorrected. Infrared (IR) spectra (KBr) were recorded on a Jasco FT/IR-4200 instrument. ^{1}H NMR (400 MHz) and ^{13}C NMR (100 MHz) on a Brucker AV 400 MHz spectrometer were recorded in DMSO-d_6 or $CDCl_3$. Chemical shifts are reported in δ (ppm) units relative to the internal standard tetramethylsilane (TMS). Mass spectra were obtained on an Agilent MS/5975 mass spectrometer. Elemental analyses were performed on a CARLOERBA 1106 instrument and the results of elemental analyses for C, H, N, and S were within ±0.4% of the theoretical values. All chemicals and solvents used were of reagent grade and were purified and dried by standard methods before use. All air-sensitive reactions were run under a nitrogen atmosphere. All the reactions were monitored by thin-layer chromatography (TLC) on pre-coated silica gel G plates at 254 nm under a UV lamp using ethyl acetate/hexane as eluents. Flash chromatography separations were obtained on silica gel (300–400 mesh). Compounds **6a–6d** and **6f–6i** were obtained commercially; 3,5-dimethylphenyl acetic acid (**6e**) was prepared according to literature procedure.[16,17]

5.1.2. General procedure for the preparation of β-ketoesters 7a–p. To a well stirred solution of substituted diethyl malonate (517 mmol) in anhydrous EtOH (345 mL) was added dropwise a solution of KOH (29 g, 517 mmol) in EtOH (345 mL) at room temperature over 4 h. Then the resulting mixture was allowed to stand at this temperature for four to twelve hours until the pH of the final mixture had a value between 7 and 8. After removing the solvent, the residue was rinsed with Et_2O and suspended in anhydrous CH_3CN (800 mL), Et_3N (100 mL, 717 mmol) and $MgCl_2$ (54 g, 567 mmol) were added and the mixture continued being stirred at room temperature for 2 h. Then a solution of arylacetyl imidazolide, which was prepared from arylacetic acid (214 mmol) and *N,N′*-carbonyldiimidazole (CDI, 45 g, 259 mmol) in CH_3CN (400 mL) 30 min before, was added and the reaction mixture was stirred overnight at room temperature. After refluxing for 2 h and then cooled to 0 °C, a solution of 13 % HCl (800 mL) was added slowly and the resulting clear

mixture was stirred for further 15 min. The organic phase was separated and concentrated, and the aqueous phase was extracted with EtOAc (3 × 300 mL). The combined organic layers were washed with saturated $NaHCO_3$ (3 × 350 mL) and brine (3 × 350 mL), dried (Mg_2SO_4), filtered and concentrated in vacuo to afford the crude products **7a–p** to be used directly in the next step without further purification.

5.1.3. General procedure for the preparation of 5-alkyl-6-substituted thiouracil 8a–p. To a stirred solution of EtONa (250 mmol) in EtOH (30 mL) was added thiourea (15.3 g, 200 mmol) at room temperature. After stirring for 30 min, the corresponding β-ketoesters **7a–p** (160 mmol) was added and the reaction mixture was refluxed for 16 h under a nitrogen atmosphere. The reaction mixture was cooled to room temperature and concentrated under reduced pressure, and the remaining residue was dissolved in H_2O and neutralized with 13% HCl to precipitate. The solid was collected, washed sequentially with H_2O, EtOH, and Et_2O, then dried to give **8a–p** to be used in the next step without further purification.

5.1.4. General procedure for the preparation of target compounds 5a–q. To a stirred solution of **8a–p** (3 mmol) in anhydrous DMF (18 mL) was added K_2CO_3 (0.46 g, 3.3 mmol) at room temperature. After stirring for 20 min, appropriate arylcarbonylmethyl halide (3.3 mmol) was added, and stirring was continued at this temperature for another 18 h. The reaction mixture was poured into cold H_2O (150 mL), the resulting precipitate was collected by filtration and washed sequentially with small portions of H_2O, MeOH and Et_2O and then dried in vacuo at 40 °C to afford the corresponding crude product, which was purified by flash chromatography or by crystallization to give the pure target compounds **5a–q**.

5.1.4.1. 6-(2-Bromobenzyl)-2-(4-methoxyphenylcarbonylmethylthio)-5-methylpyrimidin-4(3*H*)-one (5a). Yield 21%; recrystallized from MEK as a pale yellow crystal; mp 204.3–205.2 °C; FT-IR (KBr) *ν* 3407 (NH), 2958 (CH_3), 2917, 2841 (CH_2), 1671 (C=O), 1658 (C=O), 1597, 1513, 1458 (aryl); ^{1}H NMR (DMSO-d_6) *δ* 1.94 (s, 3H, CH_3), 3.82 (s, 2H, ArCH_2), 3.87 (s, 3H, OCH_3), 4.50 (s, 2H, SCH_2), 7.02 (d, 2H, *J* = 8.8 Hz, Ar′$H_{3,5}$), 7.04–7.12 (m, 3H, Ar$H_{4,5,6}$), 7.44 (d, 1H, ArH_3), 7.80 (d, 2H, *J* = 8.8 Hz, Ar′$H_{2,6}$), 12.73 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) *δ* 10.45 (*C*H_3), 37.61 (Ar*C*H_2), 40.04 (S*C*H_2), 55.72 (O*C*H_3), 113.97 (2C), 115.61 (C-5), 124.30, 127.44, 128.35, 128.56, 130.60 (2C), 131.40, 132.22, 137.51, 156.48 (C-6), 159.15 (C-2), 163.36 (C-4), 163.53 (Ar′-C_4), 191.23 (C=O); MS (EI) *m/z* 458 (M^+); Anal. calcd for $C_{21}H_{19}BrN_2O_3S$: C, 54.91; H, 4.17; N, 6.10; S, 6.98. Found: C, 54.89; H, 4.18; N, 6.11; S, 6.96.

5.1.4.2. 6-(3-Bromobenzyl)-2-(4-methoxyphenylcarbonylmethylthio)-5-methylpyrimidin-4(3*H*)-one (5b). Yield 70%; column chromatography (eluent CH_2Cl_2/AcOEt 100:20), white plate crystal; mp 180.8–181.3 °C; FT-IR (KBr) *ν* 3428 (NH), 2935, 2910, 2839 (CH_3, CH_2), 1666 (C=O), 1638 (C=O), 1595, 1570, 1548, 1476 (aryl); ^{1}H NMR (DMSO-d_6) *δ* 1.91 (s, 3H, CH_3), 3.66 (s, 2H, ArCH_2), 3.86 (s, 3H, OCH_3), 4.64 (s, 2H, SCH_2), 7.02–7.11 (m, 4H, Ar$H_{5,6}$ and Ar′$H_{3,5}$), 7.26 (s, 1H, ArH_2), 7.32 (d, 1H, *J* = 7.6 Hz ArH_4), 7.96 (d, 2H, *J* = 8.8 Hz, Ar′$H_{2,6}$]), 12.73 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) *δ* 10.47 (*C*H_3), 37.36 (Ar*C*H_2), 39.60 (S*C*H_2), 55.75 (O*C*H_3), 114.09 (2C), 116.25 (C-5), 121.63, 128.00, 128.86, 129.24, 130.38, 130.74 (2C), 131.42, 140.85, 155.82 (C-6), 159.27 (C-2), 162.97 (C-4), 163.52 (Ar′-C_4), 191.53 (C=O); MS (EI) *m/z* 458 (M^+); Anal. calcd for $C_{21}H_{19}BrN_2O_3S$: C, 54.91; H, 4.17; N, 6.10; S, 6.98. Found: C, 54.95; H, 4.14; N, 6.12; S, 7.01.

5.1.4.3. 6-(3-Hydroxybenzyl)-2-(4-methoxyphenylcarbonylmethylthio)-5-methylpyrimidin-4(3*H*)-one (5c). Yield 64%; recrystallized from AcOEt as an off-white powder; mp 185.0–185.4 °C; FT-IR (KBr) *ν* 3447 (NH, OH), 2965 (CH_3), 2919, 2840 (CH_2), 1644 (C=O), 1598, 1544, 1509, 1456 (aryl); ^{1}H NMR (DMSO-d_6) *δ* 1.82 (s, 3H, CH_3), 3.57 (s, 2H, ArCH_2), 3.83 (s, 3H, OCH_3), 4.65 (s, 2H, SCH_2), 6.45 (d, 1H, *J* = 7.6 Hz, ArH_2), 6.56 (m, 2H, Ar$H_{5,6}$), 6.95 (t, 1H, J_1 = 7.6 Hz, J_2 = 7.6 Hz, ArH_4), 7.03 (d, 2H, *J* = 8.8 Hz, Ar′$H_{3,5}$), 7.96 (d, 2H, *J* = 8.8 Hz, Ar′$H_{2,6}$), 9.24 (br s, 1H, ArO*H*), 12.54 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) *δ*10.51 (*C*H_3), 37.27 (Ar*C*H_2), 55.70 (O*C*H_3), 113.29, 114.08 (2C), 115.14 (C-5), 115.55, 119.55, 128.88, 129.24, 130.75 (2C), 139.35, 156.53 (C-6), 157.40 (Ar-C_3), 160.99 (C-2), 163.51 (2C, C-4 and Ar′-C_4), 191.87 (C=O); MS (EI) *m/z* 396 (M^+); Anal. calcd for $C_{21}H_{20}N_2O_4S$: C, 63.62; H, 5.08; N, 7.07; S, 8.09. Found: C, 63.65; H, 5.04; N, 7.05; S, 8.06.

5.1.4.4. 6-(4-Bromobenzyl)-2-(4-methoxyphenylcarbonylmethylthio)-5-methylpyrimidin-4(3*H*)-one (5d). Yield 17%; recrystallized from THF as a white solid; mp 199.2–199.6 °C; FT-IR (KBr) *ν* 3433 (NH), 2954 (CH_3), 2919, 2835 (CH_2), 1667 (C=O), 1641 (C=O), 1595, 1573, 1547, 1457 (aryl); ^{1}H NMR (DMSO-d_6) *δ* 1.98 (s, 3H, CH_3), 3.68 (s, 2H, ArCH_2), 3.92 (s, 3H, OCH_3), 4.71 (s, 2H, SCH_2), 7.04 (d, 2H, *J* = 8.4 Hz, Ar$H_{2,6}$), 7.12 (d, 2H, *J* = 8.8 Hz, Ar′$H_{3,5}$), 7.33 (d, 2H, *J* = 8.4 Hz, Ar$H_{3,5}$), 8.03 (d, 2H, *J* = 8.8 Hz, Ar′$H_{2,6}$); ^{13}C NMR (DMSO-d_6) *δ* 10.43 (*C*H_3), 37.38 (Ar*C*H_2), 39.36 (S*C*H_2), 55.71 (O*C*H_3), 114.09 (2C), 115.25 (C-5), 119.39, 128.84, 130.68 (2C), 131.06 (4C), 137.49, 156.70 (C-6), 160.09 (C-2), 163.45 (C-4), 163.52 (Ar′-C_4), 191.43 (C=O); MS (EI) *m/z* 458 (M^+); Anal. calcd for $C_{21}H_{19}BrN_2O_3S$: C, 54.91; H, 4.17; N, 6.10; S, 6.98. Found: C, 54.95; H, 4.18; N, 6.04; S, 6.96.

5.1.4.5. 6-(3,5-Dimethylbenzyl)-2-(4-methoxyphenylcarbonylmethylthio)-5-methylpyrimidin-4(3*H*)-one (5e). Yield 38%; column chromatography (eluent CH_2Cl_2/AcOE 100:20), off-white plate crystal; mp 197.3–197.7 °C; FT-IR (KBr) *ν* 3433 (NH), 2950 (CH_3), 2914, 2848 (CH_2), 1673 (C=O), 1655 (C=O), 1599, 1573, 1554, 1509, 1457 (aryl); ^{1}H NMR ($CDCl_3$) *δ* 1.90 (s, 3H, CH_3), 2.13 (s, 6H, Ar(CH_3)$_2$), 3.56 (s, 2H, ArCH_2), 3.84 (s, 3H, OCH_3), 4.67 (s, 2H, SCH_2), 6.63 (s, 2H, Ar$H_{2,6}$), 6.74 (s, 1H, ArH_4), 7.04 (d, 2H,

J = 8.8 Hz, Ar′$H_{3,5}$), 8.00 (d, 2H, J = 8.8 Hz, Ar′$H_{2,6}$), 12.66 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 10.50 (CH$_3$), 20.93 (2C, Ar(CH$_3$)$_2$), 37.22 (ArCH$_2$), 55.68 (OCH$_3$), 114.04 (2C), 115.01 (C-5), 126.45 (2C), 127.73, 128.99, 130.66 (2C), 137.21 (2C), 137.83, 156.48 (C-6), 160.84 (C-2), 163.47 (2C, C-4 and Ar′-C$_4$), 191.69 (C=O); MS (EI) m/z 408 (M$^+$); Anal. calcd for $C_{23}H_{24}N_2O_3S$: C, 67.62; H, 5.92; N, 6.86; S, 7.85. Found: C, 67.60; H, 5.95; N, 6.88; S, 7.81.

5.1.4.6. 6-(3,5-Bis(trifluoromethyl)benzyl)-2-(4-methoxyphenylcarbonylmethylthio)-5-methylpyrimid in-4(3*H*)-one (5f). Yield 35%; recrystallized from AcOEt as a white needle crystal; mp 202.8–203.1 °C; FT-IR (KBr) v 3397 (NH), 2932, 2894, 2848 (CH$_3$, CH$_2$), 1653 (C=O), 1602 (C=O), 1572, 1552, 1509 (aryl); ^{1}H NMR (DMSO-d_6) δ 1.93 (s, 3H, CH_3), 3.81 (s, 3H, OCH_3), 3.94 (s, 2H, ArCH_2), 4.51 (s, 2H, SCH_2), 6.94 (d, 2H, J = 8.8 Hz, Ar′$H_{3,5}$), 7.74–7.77 (m, 3H, ArH), 7.78 (d, 2H, J = 8.8 Hz, Ar′$H_{2,6}$), 12.76 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 10.37 (CH$_3$), 37.11 (ArCH$_2$), 38.99 (SCH$_2$), 55.65 (OCH$_3$), 113.88 (2C), 115.09 (C-5), 120.00 (sept, J = 4 Hz, Ar-C$_4$), 123.45 (q, 2C, J = 271.1 Hz, CF$_3$), 128.52, 129.83 (2C), 129.99 (q, 2C, J = 32.5 Hz, Ar-C$_{3,5}$), 130.42 (2C), 141.60, 155.06 (C-6), 158.20 (C-2), 162.77 (C-4), 163.45 (Ar′-C$_4$), 191.12 (C=O); MS (EI) m/z 516 (M$^+$); Anal. calcd for $C_{23}H_{18}F_6N_2O_3S$: C, 53.49; H, 3.51; N, 5.42; S, 6.21. Found: C, 53.53; H, 3.46; N, 5.40; S, 6.22.

5.1.4.7. 6-(2,6-Dichlororbenzyl)-2-(4-methoxyphenylcarbonylmethylthio)-5-methylpyrimidin-4(3*H*)-one (5g). Yield 52%; recrystallized from dioxone as a white block crystal; mp 230.5–235.6 °C; FT-IR (KBr) v 3430 (NH), 2962 (CH$_3$), 2927, 2842 (CH$_2$), 1657 (C=O), 1596, 1569, 1509, 1434 (aryl); ^{1}H NMR (DMSO-d_6) δ 2.03 (s, 3H, CH_3), 3.90 (s, 3H, OCH_3), 4.08 (s, 2H, ArCH_2), 4.39 (s, 2H, SCH_2), 7.02–7.15 (m, 5H, Ar$H_{3,4,5}$ and Ar$H'_{3,5}$), 7.73 (d, 2H, J = 8.4 Hz, Ar$H'_{2,6}$), 12.69 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 10.16 (CH$_3$), 35.25 (ArCH$_2$), 38.02 (SCH$_2$), 55.81 (OCH$_3$), 114.02 (2C), 115.19 (C-5), 127.82 (2C), 128.10, 128.83, 130.60 (2C), 134.48, 135.40 (2C), 156.38 (C-6), 157.88 (C-2), 163.17 (C-4), 163.69 (Ar′-C$_4$), 190.28 (C=O); MS (EI) m/z 448 (M$^+$); Anal. calcd for $C_{21}H_{18}Cl_2N_2O_3S$: C, 56.13; H, 4.04; N, 6.23; S, 7.14. Found: C, 56.10; H, 4.08; N, 6.21; S, 7.14.

5.1.4.8. 6-(α-Phenylbenzyl)-2-(4-methoxyphenylcarbonylmethylthio)-5-methylpyrimidin-4(3*H*)-one (5h). Yield 26%; recrystallized from THF as white solid; mp 220.5–221.6 °C; FT-IR (KBr) v 3419 (NH), 2912, 2841 (CH$_3$, CH$_2$), 1671 (C=O), 1636 (C=O), 1601, 1574, 1542, 1510, 1493, 1423 (aryl); ^{1}H NMR (DMSO-d_6) δ 1.93 (s, 3H, CH_3), 3.85 (s, 3H, OCH_3), 4.52 (s, 2H, SCH_2), 5.49 (s, 1H, ArCH), 7.04–7.12 (m, 12H, 2 × ArH and Ar′$H_{3,5}$), 7.87 (d, 2H, J = 9.2 Hz Ar′$H_{2,6}$), 12.77 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 10.16 (CH$_3$), 38.03 (SCH$_2$), 53.17 (ArCH), 55.74 (OCH$_3$), 114.05 (2C), 115.64 (C-5), 126.41 (2C), 128.04 (4C), 128.47, 129.19 (4C), 130.73 (2C), 141.49 (2C), 156.31 (C-6), 161.86 (C-2), 163.60 (2C, C-4 and Ar′-C$_4$), 190.75 (C=O); MS (EI) m/z 456 (M$^+$); Anal. calcd for $C_{27}H_{24}N_2O_3S$: C, 71.03; H, 5.30; N, 6.14; S, 7.02. Found: C, 71.08; H, 5.28; N, 6.12; S, 7.05.

5.1.4.9. 5-Methyl-6-(1-naphthylmethyl)-2-(phenylcarbonylmethylthio)pyrimidin-4(3*H*)-one (5i). Yield 21%; recrystallized from dioxone as a white solid; mp 194.3–195.3 °C; FT-IR (KBr) v 3432 (NH), 2928, 2840 (CH$_3$, CH$_2$), 1690 (C=O), 1633 (C=O), 1595, 1579, 1553, 1509, 1480, 1448 (aryl); ^{1}H NMR (DMSO-d_6) δ 1.07 (s, 3H, CH$_3$), 4.14 (s, 2H, CH$_2$-naphthyl), 4.48 (s, 2H, SCH_2), 7.10–7.94 (m, 12H, aryl), 12.72 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 10.57 (CH$_3$), 37.45 (CH$_2$-naphthyl), 37.75 (SCH$_2$), 115.66 (C-5), 124.13–135.81 (16C, aryl), 156.50 (C-6), 160.63 (C-2), 163.54 (C-4), 193.18 (C=O); MS (EI) m/z 400 (M$^+$); Anal. calcd for $C_{24}H_{20}N_2O_2S$: C, 71.98; H, 5.03; N, 6.99; S, 8.01. Found: C, 71.95; H, 5.07; N, 7.03; S, 7.99.

5.1.4.10. 6-(2-Bromobenzyl)-5-ethyl-2-(4-methoxyphenylcarbonylmethylthio)pyrimidin-4(3*H*)-one (5j). Yield 62%; recrystallized from THF as white solid; mp 213.2–213.8 °C; FT-IR (KBr) v 3420 (NH), 2962 (CH$_3$), 2926, 2845 (CH$_2$), 1649 (C=O), 1601 (C=O), 1570, 1546, 1509, 1422 (aryl); ^{1}H NMR (DMSO-d_6) δ 0.97 (t, 3H, J = 7.2 Hz, CH$_2$CH_3), 2.42 (q, 2H, J = 7.2 Hz, CH_2CH$_3$), 3.84 (s, 2H, ArCH_2), 3.87 (s, 3H, OCH_3), 4.49 (s, 2H, SCH_2), 7.01 (d, 2H, J = 8.8 Hz, Ar′$H_{2,6}$), 7.05–7.13 (m, 3H, Ar$H_{4,5,6}$), 7.43 (d, 1H,, J = 7.6 Hz, ArH_3), 7.80 (d, 2H, J = 8.8 Hz, Ar′$H_{2,6}$), 12.70 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 12.30 (CH$_2$$CH_3$), 18.25 ($CH_2CH_3$), 37.55 (Ar$CH_2$), 55.76 (O$CH_3$), 113.99 (2C), 121.86 (C-5), 124.34, 127.47, 128.39, 128.56, 130.64 (2C), 131.39, 132.23, 137.73, 155.87 (C-6), 158.39 (C-2), 162.59 (C-4), 163.53 (Ar′-C$_4$), 191.26 (C=O); MS (EI) m/z 472 (M$^+$); Anal. calcd for $C_{22}H_{21}BrN_2O_3S$: C, 55.82; H, 4.47; N, 5.92; S, 6.77. Found: C, 55.86; H, 4.45; N, 5.90; S, 6.78.

5.1.4.11. 6-(3-Bromobenzyl)-5-ethyl-2-(4-methoxyphenylcarbonylmethylthio)pyrimidin-4(3*H*)-one (5k). Yield 47%; column chromatography (eluent CH$_2$Cl$_2$/AcOE 100:20), white needle crystal; mp 201.9–203.0 °C; FT-IR (KBr) v 3431 (NH), 2967 (CH$_3$), 2924, 2840 (CH$_2$), 1652 (C=O), 1600 (C=O), 1570, 1550, 1510, 1423 (aryl); ^{1}H NMR (DMSO-d_6) δ 0.91 (t, 3H, J = 7.2 Hz, CH$_2$CH_3), 2.41 (q, 2H, J = 7.2 Hz, CH_2CH$_3$), 3.67 (s, 2H, ArCH_2), 3.85 (s, 3H, OCH_3), 4.64 (s, 2H, SCH_2), 7.03–7.09 (m, 4H, Ar$H_{5,6}$ and Ar′$H_{3,5}$), 7.28 (s, 1H, ArH_2), 7.32 (d, 1H, ArH_4), 7.96 (d, 2H, J = 8.8 Hz, Ar′$H_{2,6}$), 12.72 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 13.21 (CH$_2$$CH_3$), 18.25 ($CH_2CH_3$), 37.38 (Ar$CH_2$), 55.74 (O$CH_3$), 114.08 (2C), 121.59, 121.93 (C-5), 127.99, 128.84, 129.21, 130.34, 130.73 (2C), 131.45, 141.21, 155.97 (C-6), 159.19 (C-2), 162.67 (C-4), 163.51 (Ar′-C$_4$), 191.50 (C=O); MS (EI) m/z 472 (M$^+$); Anal. calcd for $C_{22}H_{21}BrN_2O_3S$: C, 55.82; H, 4.47; N, 5.92; S, 6.77. Found: C, 55.80; H, 4.51; N, 5.89; S, 6.81.

5.1.4.12. 6-(4-Bromobenzyl)-5-ethyl-2-(4-methoxyphenylcarbonylmethylthio)pyrimidin-4(3*H*)-one (5l). Yield 58%; column chromatography (eluent CH$_2$Cl$_2$/AcOEt 100:20), white solid; mp 205–206.7 °C; FT-IR (KBr) v 3434 (NH), 2963 (CH$_3$), 2928 (CH$_2$), 2870 (CH$_3$), 2836

(CH_2), 1666 (C=O), 1638 (C=O), 1593, 1571, 1543, 1457 (aryl); ^{1}H NMR (DMSO-d_6) δ 0.98 (t, 3H, J = 7.2 Hz, CH_2CH_3), 2.46 (q, 2H, J = 7.2 Hz, CH_2CH_3), 3.69 (s, 2H, $ArCH_2$), 3.91 (s, 3H, OCH_3), 4.69 (s, 2H, SCH_2), 7.04 (d, 2H, J = 8.0 Hz, $ArH_{2,6}$), 7.11 (d, 2H, J = 8.8 Hz, $Ar'H_{3,5}$), 7.31 (d, 2H, J = 8.0 Hz, $ArH_{3,5}$), 8.00 (d, 2H, J = 8.8 Hz, $Ar'H_{2,6}$), 12.73 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 13.35 (CH_2CH_3), 18.26 (CH_2CH_3), 37.45 ($ArCH_2$), 38.83 (SCH_2), 55.79 (OCH_3), 114.14 (2C), 119.42, 121.79 (C-5), 128.86, 130.76 (2C), 131.07 (2C), 131.17 (2C), 137.84, 156.10 (C-6), 159.12 (C-2), 162.56 (C-4), 163.56 (Ar′-C_4), 191.42 (C=O); MS (EI) m/z 472 (M^+); Anal. calcd for $C_{22}H_{21}BrN_2O_3S$: C, 55.82; H, 4.47; N, 5.92; S, 6.77. Found: C, 55.85; H, 4.43; N, 5.91; S, 6.75.

5.1.4.13. 6-(4-Phenylbenzyl)-5-ethyl-2-(4-methoxyphenylcarbonylmethylthio)pyrimidin-4(3*H*)-one (5m). Yield 30%; column chromatography, (eluent CH_2Cl_2/AcOEt 100:20), white solid; mp 201.2–201.6 °C; FT-IR (KBr) ν 3421 (NH), 2965 (CH_3), 2920 (CH_2), 2870 (CH_3), 2837 (CH_2), 1660 (C=O), 1601 (C=O), 1575, 1551, 1509 (aryl); ^{1}H NMR (DMSO-d_6) δ 0.96 (t, 3H, J = 7.2 Hz, CH_2CH_3), 2.46 (q, 2H, J = 7.2 Hz, CH_2CH_3), 3.71 (s, 2H, $ArCH_2$), 3.80 (s, 3H, OCH_3), 4.69 (s, 2H, SCH_2), 6.99–8.00 (m, 13H, aryl), 12.68 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 13.21 (CH_2CH_3), 18.20 (CH_2CH_3), 37.39 ($ArCH_2$), 55.60 (OCH_3), 114.00–140.00 (18C, aryl and C-5), 155.60 (C-6), 160.71 (C-2), 162.45 (C-4), 163.49 (Ar′-C_4), 191.46 (C=O); MS (EI) m/z 470 (M^+). Anal. calcd for $C_{28}H_{26}N_2O_3S$: C, 71.46; H, 5.57; N, 5.95; S, 6.81. Found: C, 71.43; H, 5.61; N, 5.93; S, 6.85.

5.1.4.14. 6-(3,5-Dimethylbenzyl)-5-ethyl-2-(4-methoxyphenylcarbonylmethylthio)pyrimidin-4(3*H*)-one (5n). Yield 33%; column chromatography (eluent CH_2Cl_2/AcOEt 100:20), white needle crystal; mp 201.7–202.6 °C; FT-IR (KBr) ν 3432 (NH), 2968, 2928, 2867 (CH_3, CH_2), 1653 (C=O), 1600 (C=O), 1571, 1542, 1509, 1458 (aryl); ^{1}H NMR (DMSO-d_6) δ 0.90 (t, 3H, J = 7.2 Hz, CH_2CH_3), 2.13 (s, 6H, $Ar(CH_3)_2$), 2.40 (q, 2H, J = 7.2 Hz, CH_2CH_3), 3.32 (s, 2H, $ArCH_2$), 4.67(s, 2H, SCH_2), 6.65 (s, 2H, $ArH_{2,6}$), 6.74 (s, 1H, ArH_4), 7.0 (d, 2H, J=8.8 Hz, $Ar'H_{3,5}$), 7.99 (d, 2H, J = 8.8 Hz, $Ar'H_{2,6}$), 12.68 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 13.11 (CH_2CH_3), 18.31 (CH_2CH_3), 20.93 (2C, $Ar(CH_3)_2$), 37.25 ($ArCH_2$), 55.70 (OCH_3), 114.04 (2C), 121.09 (C-5), 126.49 (2C), 127.71, 128.98, 130.67 (2C), 137.17 (2C), 138.19, 156.27 (C-6), 160.28 (C-2), 163.13 (C-4), 163.48 (Ar′-C_4), 191.66 (C=O); MS (EI) m/z 422 (M^+); Anal. calcd for $C_{24}H_{26}N_2O_3S$: C, 68.22; H, 6.20; N, 6.63; S, 7.59. Found: C, 68.18; H, 6.23; N, 6.64; S, 7.55.

5.1.4.15. 6-(3,5-Bis(trifluoromethyl)benzyl)-5-ethyl-2-(4-methoxyphenylcarbonylmethylthio)pyrimid in-4(3*H*)-one (5o). Yield 55%; column chromatography (eluent Petroleum ether/acetone 100:25), white solid; mp 172.3–173.3 °C; FT-IR (KBr) ν 3422 (NH), 2966 (CH_3), 2933, 2848 (CH_2), 1671 (C=O), 1640 (C=O), 1597, 1576, 1557, 1513, 1458 (aryl); ^{1}H NMR (DMSO-d_6) δ 0.92 (t, 3H, J = 7.4 Hz, CH_2CH_3), 2.45 (q, 2H, J = 7.2 Hz, CH_2CH_3), 3.82 (s, 3H, OCH_3), 3.97 (s, 2H, $ArCH_2$), 4.53 (s, 2H, SCH_2), 6.95 (d, J = 8.8 Hz, $Ar'H_{3,5}$), 7.76 (s, 2H, $ArH_{2,6}$), 7.78 (s, 1H, ArH_4), 7.80 (d, J = 8.8 Hz, $Ar'H_{2,6}$), 12.78 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 13.15 (CH_2CH_3), 18.15 (CH_2CH_3), 37.14 ($ArCH_2$), 38.50 (SCH_2), 55.67 (OCH_3), 113.88 (2C), 120.04 (sept, J = 4 Hz, Ar-C_4), 122.45 (C-5), 123.48 (q, 2C, J = 271.1 Hz, CF_3), 128.55, 129.84 (2C), 130.00 (q, 2 C, J = 32.3 Hz, Ar-$C_{3,5}$), 130.46 (2C), 141.93, 156.31 (C-6), 157.85 (C-2), 162.49 (C-4), 163.46 (Ar′-C_4), 191.16 (C=O); MS (EI) m/z 530 (M^+); Anal. calcd for $C_{24}H_{20}F_6N_2O_3S$: C, 54.34; H, 3.80; N, 5.28; S, 6.04. Found: C, 54.31; H, 3.84; N, 5.30; S, 6.10.

5.1.4.16. 6-(3,5-Dimethylbenzyl)-5-ethyl-2-(phenylcarbonylmethylthio)pyrimidin-4(3*H*)-one (5p). Yield 38%; column chromatography (eluent CH_2Cl_2/AcOEt 100:20), yellow solid; mp 165.5–167.0 °C; FT-IR (KBr) ν 3431 (NH), 2964 (CH_3), 2929 (CH_2), 2869 (CH_3), 1637 (C=O), 1600, 1570, 1542, 1450 (aryl); ^{1}H NMR (DMSO-d_6) δ 0.89 (t, 3H, J = 7.4 Hz, CH_2CH_3), 2.11 (s, 6H, $Ar(CH_3)_2$), 2.39 (q, 2H, J = 7.4 Hz, CH_2CH_3), 3.54 (s, 2H, $ArCH_2$), 4.71 (s, 2H, SCH_2), 6.61 (s, 2H, $ArH_{2,6}$), 6.71 (s, 1H, ArH_4), 7.52 (t, 2H, $Ar'H_{3,5}$), 7.65 (t, 1H, $Ar'H_4$), 7.90 (d, 2H, $Ar'H_{2,6}$), 12.69 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 13.14 (CH_2CH_3), 18.33 (CH_2CH_3), 20.95 (2C, $Ar(CH_3)_2$), 37.65 ($ArCH_2$), 121.19 (C-5), 126.48 (2C), 127.72, 128.31 (2C), 128.86 (2C), 133.56, 136.24, 137.17 (2C), 138.17, 156.00 (C-6), 160.35 (C-2), 163.04 (C-4), 193.39 (C=O); MS (EI) 392 (M^+); Anal. calcd for $C_{23}H_{24}N_2O_2S$: C, 70.38; H, 6.16; N, 7.14; S, 8.17. Found: C, 70.33; H, 6.19; N, 7.13; S, 8.21.

5.1.4.17. 5-Ethyl-2-(4-methoxyphenylcarbonylmethylthio)-6-(α-phenylbenzyl)pyrimidin-4(3*H*)-one (5q). Yield 12%; column chromatography (eluent CH_2Cl_2/AcOEt 100:10), white solid; mp 214.3–214.9 °C; FT-IR (KBr) ν 3397 (NH), 2962 (CH_3), 2916 (CH_2), 2869 (CH_3), 1672 (C=O), 1637 (C=O), 1600, 1574, 1543, 1494, 1457 (aryl); ^{1}H NMR (DMSO-d_6) δ 0.94 (t, 3H, J = 7.2 Hz, CH_2CH_3), 2.54 (q, 2H, J = 7.2 Hz, CH_2CH_3), 3.91 (s, 3H, OCH_3), 4.60 (s, 2 H, $ArCH_2$), 5.56 (s, 1H, ArH), 7.11–7.20 (m, 12H, 2 × ArH and $Ar'H_{3,5}$), 7.95 (d, 2H, J = 8.8 Hz, $Ar'H_{2,6}$), 12.82 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6) δ 13.37 (CH_2CH_3), 18.01 (CH_2CH_3), 37.99 (SCH_2), 52.58 ($ArCH$), 55.73 (OCH_3), 114.04 (2C), 121.63 (C-5), 126.39 (2C), 128.02 (4C), 128.47, 129.15 (4C), 130.72 (2C), 141.84 (2C), 156.16 (C-6), 161.16 (C-2), 163.09 (C-4), 163.58 (Ar′-C_4), 190.68 (C=O); MS (EI) m/z 470 (M^+); Anal. calcd for $C_{28}H_{26}N_2O_3S$: C, 71.46; H, 5.57; N, 5.95; S, 6.81. Found: C, 71.41; H, 5.59; N, 5.89; S, 6.83.

5.2. Anti-HIV activity assays

The activity of the compounds against wild-type HIV-1, the double RT mutant virus strain (RES056) and HIV-2 (ROD) was based on the inhibition of a virus-induced cytopathic effect in MT-4 cells using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) method.[18] Briefly, virus stocks were titrated in MT-4 cells and expressed as 50% cell culture infective dose

($CCID_{50}$). MT-4 cells were suspended in culture medium at 1×10^5 cells/mL and infected with HIV at a multiplicity of infection of 0.02. Immediately after virus infection, 100 μL of the cell suspension was brought into each well of a flat-bottomed microtiter tray containing various concentrations of the test compounds. The test compounds were dissolved in DMSO at 50 mM. After a 4-day incubation at 37 °C, the number of viable cells was determined using the MTT method. Compounds were tested in parallel for cytotoxic effect in uninfected MT-4 cells.

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