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Original article

Synthesis and in vitro anti-HIV evaluation of a new series of 6-arylmethyl-substituted *S*-DABOs as potential non-nucleoside HIV-1 reverse transcriptase inhibitors

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ABSTRACT

A series of new 5-alkyl-2-benzylsulfanylpyrimidin-4(3*H*)-ones (**5a**–**y**) bearing different substituted arylmethyl moieties at the C-6 position of the pyrimidine core have been synthesized and evaluated for their in vitro activities against HIV-1 and HIV-2 in MT-4 cell cultures. The majority of the title compounds showed moderate to good activities against HIV-1 with an IC_{50} range from 6.67 μM to 0.12 μM. Among them, 6-(3,5-dimethylbenzyl) analogue **5q** exhibited the most potent anti-HIV-1 activity ($IC_{50} = 0.12$ μM, $SI > 2642$), which was about 40-fold more active than the reference compounds 1-[(2-hydroxyethoxy)methyl]-6-(phenylsulfanyl)thymine (HEPT) and 2′,3′-dideoxyinosine (DDI). The structure–activity relationships (SARs) of these new congeners were further discussed.

1. Introduction

In our continuing efforts to find novel effective and selective anti-HIV-1 agents in 2-alkylsulfanyl-6-benzyl-3,4-dihydropyrimidin-4(3*H*)-ones (*S*-DABOs) type NNRTIs [1] (**1**, Fig. 1), we have synthesized a series of 2-arylcarbonylmethysulfanyl-substituted *S*-DABOs [2–4] (**2**, Fig. 1), many of which exhibited potent anti-HIV-1 activity and high selectivity index (SI) comparable to the well-known 2-alkylsulfanyl-substituted *S*-DABOs [5] (F_2-*S*-DABOs, **3**, Fig. 1). The follow-up structure–activity relationship (SAR) studies on these analogues revealed that the C-2, C-5 and C-6 substituent effects were tightly linked: the optimal moieties at positions 5 and 6 of the pyrimidine nucleus were dependent on the nature of the C-2 side chain. For example, there seems to be a general trend of increasing anti-HIV-1 activity (*i*-Pr > Et > Me) in 2-arylcarbonylmethylsulfanyl-substituted *S*-DABOs [2]; while in 2-alkylsulfanyl-substituted *S*-DABOs, the steric bulkiness of the C-5 substituent is detrimental to HIV-1 inhibitory activity (H ≥ Me > Et > i-Pr) [6]. Very recently, Botta et al. [7] disclosed a new series of 2-arylalkylsulfanyl-substituted *S*-DABOs (**4**, Fig. 1) with a lead compound **4a** displaying potent anti-HIV-1 activity both in enzyme and cell-based assays. Inspired by these promising results and based on the above-mentioned SAR, a new series of 5-alkyl-6-arylmethyl-2-benzylsulfanyl-substituted *S*-DABOs (**5**, Fig. 1) were synthesized in hopes of further exploration of the structure–activity relationships of *S*-DABOs and discovery of new more potent HIV-1 inhibitors. In this paper, we described the synthesis, anti-HIV-1 activity and preliminary SAR studies of these new congeners.

2. Results and discussion

2.1. Chemistry

The synthetic route to the target compounds **5a**–**y** is depicted in Scheme 1. The key intermediate β-ketoesters **7a**–**n** were easily

Abbreviations: CDI, 1,1′-carbonyldiimidazole; DABO, 2-alkoxyl-6-benzyl-3,4-dihydropyrimidin-4(3*H*)-ones; DDI, 2′,3′-dideoxyinosine; DMF, *N*,*N*-dimethylformamide; HEPT, 1-[(2-hydroxyethoxy)methyl]-6-(phenylsulfanyl)thymine; NNRTIs, non-nucleoside reverse transcriptase inhibitors; HIV, human immunodeficiency virus; HIV-1, human immunodeficiency virus type-1; HIV-2, human immunodeficiency virus type-2; MTT, 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide; *S*-DABOs, 2-alkylsulfanyl-6-benzyl-3,4-dihydropyrimidin-4(3*H*)-ones; SARs, structure–activity relationships.

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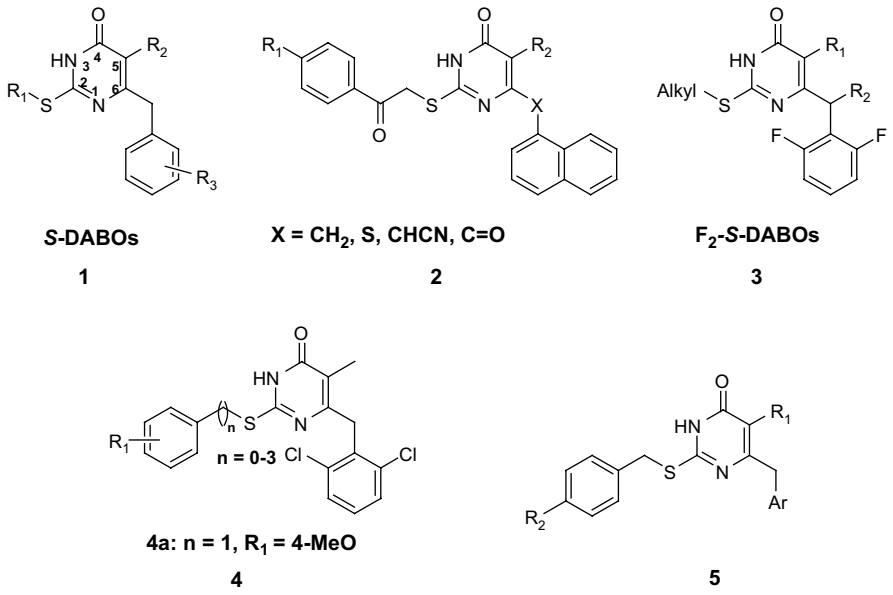


Fig. 1. Chemical structures of *S*-DABOs.

prepared in 48–98% yield using the method of Clay et al. [8] by exposure of substituted arylacetic acids **6a**–**g** to 1,1′-carbonyldiimidazole (CDI) followed by treatment with different ethyl potassium malonates in the presence of anhydrous $MgCl_2$ and Et_3N. On the other hand, the rest of β-ketoesters **7o** and **p** were prepared from commercially available 1-naphthylacetonitrile and ethyl 2-bromoalkanoate following the procedure described previously [9]. Then, condensation of **7a**–**p** with thiourea in the presence of EtONa in refluxing EtOH led to substituted uracil **8a**–**p** in 50–88% yield. Subsequent treatment of **8a**–**p** with appropriate benzyl halides in the presence of K_2CO_3 in anhydrous DMF afforded the corresponding target compounds **5a**–**y** in 14–77% yield.

All the target compounds **5a**–**y** were characterized by NMR, MS, IR and elemental analyses. Both analytical and spectral data of all the compounds are in full agreement with the proposed structures. Moreover, the X-ray crystallography of the compound **5v** [10] further confirmed the above structures.

2.2. Anti-HIV activity

The activity and cytotoxicity of these newly synthesized *S*-DABO analogues were evaluated in MT-4 cells for their ability to inhibit HIV-1- and HIV-2-induced cytopathogenicities. The results, expressed as CC_{50}, IC_{50} and SI, are illustrated in Table 1 together with those of HEPT and DDI as reference standards. In general, most of the tested compounds were noncytotoxic for MT-4 cells at doses as high as 165 μM, and only two compounds showed CC_{50} values at concentrations around 100 μM or lower (**5f** and **y**). Notably, the majority of these compounds exhibited moderate to good activities against HIV-1 with an IC_{50} range from 6.67 μM to 0.12 μM. Among them, 6-(3,5-dimethylbenzyl)-5-ethyl-2-(4-methoxybenzylsulfanyl)-pyrimidin-4(3*H*)-one (**5q**) turned out to be the most promising compound with an IC_{50} value of 0.12 μM being about 40-fold more active than the reference compounds HEPT and DDI. Some other compounds **5r**, **v**, **y**, **k** and **m**, also showed high HIV-1 inhibitory activities ($IC_{50} = 0.16$ μM, 0.16 μM, 0.21 μM, 0.23 μM and 0.28 μM, respectively) and good selectivity indices (SI = 1906, 1793, 178, 717 and 718, respectively).

As shown in Table 1, the substitution pattern of the C-6 benzyl moiety of *S*-DABOs played a determinant role for their HIV-1 inhibitory potency. In the case of monosubstituted anlaogues (**5a**–**f** and **5k**–**p**), a clear positional preference for substitution on the phenyl ring portion of the benzyl group was observed by direct comparison: the *ortho*-substituted analogues (**5a**, **b**, **k** and **l**) were more active than the corresponding *meta*-substituted ones (**5c**, **d**, **m** and **n**). Furthermore, the introduction of a bromine atom (**5e** and **o**) or a phenyl group (**5f** and **p**) into the *para*-position of the C-6 phenyl ring resulted in complete loss of inhibitory activity against HIV-1. Interestingly, in the group of disubstituted analogues (**5g**–**j** and **5q**–**w**), the most potent compounds turned out to be the 6-(3,5-dimethylbenzyl) derivatives followed by the 6-(2,6-dichlorobenzyl) analogues (**5q** vs **v** and **5r** vs **w**); and the 6-(3,5-trifluoromethylbenzyl) congeners were found to be less potent (**5t**) or devoid of any anti-HIV-1 activity (**5i**, **j** and **u**).

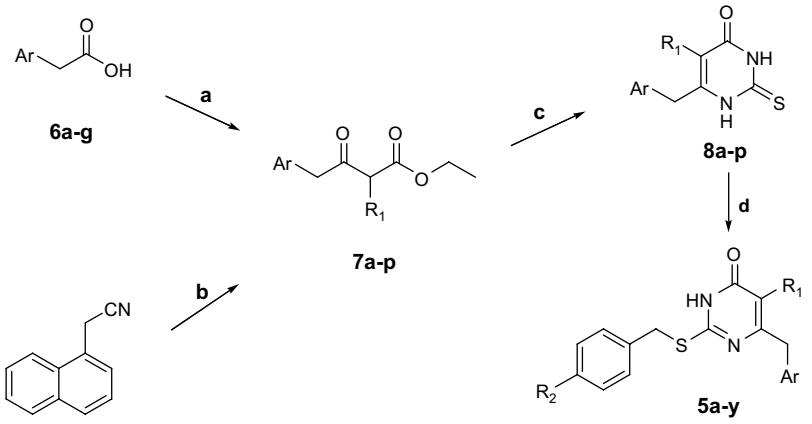
Moreover, it was found that the nature of the substituent at the C-5 position of the pyrimidine ring also influenced the anti-HIV-1 activity of these new congeners. When the C-5 substituents were changed from Me to Et or Et to i-Pr, a marked increase of anti-HIV-1 activity was observed for all the title compounds. This conclusion is in agreement with the earlier SAR in related *S*-DABO series (i.e., 2-arylcarbonylmethylsulfanyl-substituted *S*-DABOs, **2**) [2].

As for the substitution on the C-2 benzylsulfanyl group, replacement of the *para*-hydrogen (**5s**) with a methoxy group (**5q**) or a nitro group (**5r**) enhanced the HIV-1 inhibitory activity about eight and sixfold, respectively. Furthermore, the data in Table 1 indicated that the 4-methoxylbenzylsulfanyl moiety was the optimal substituent at the C-2 position of this new series of *S*-DABOs (compare **5q** vs **r**, **5v** vs **w** and **5m** vs **n**, etc.).

In addition, all the title compounds were also evaluated for their capability to inhibit the HIV-2 replication in MT-4 cells, but none was found effective (Table 1). These findings showed that this new series of *S*-DABOs was specific for HIV-1 and belonged to typical NNRTIs.

3. Conclusion

In summary, we have described the synthesis and anti-HIV-1 activity of a new series of 5-alkyl-6-arylmethyl-2-benzylsulfanyl-substituted *S*-DABOs. The bioassay results revealed that the majority of the title compounds exhibited potent HIV-1 inhibitory



6,7,8	Ar	R_1	7,8	Ar	R_1
a	2-Br-Ph	Me	**i**	3-Br-Ph	Et
b	3-Br-Ph	Me	**j**	4-Br-Ph	Et
c	4-Br-Ph	Me	**k**	4-Ph-Ph	Et
d	4-Ph-Ph	Me	**l**	2,6-Cl_2-Ph	Et
e	2,6-Cl_2-Ph	Me	**m**	3,5-Me_2-Ph	Et
f	3,5-Me_2-Ph	Me	**n**	3,5-$(CF_3)_2$-Ph	Et
g	3,5-$(CF_3)_2$-Ph	Me	**o**	1-naphthyl	Et
h	2-Br-Ph	Et	**p**	1-nahphtyl	*i*-Pr

5	Ar	R_1	R_2	5	Ar	R_1	R_2
a	2-Br-Ph	Me	MeO	**n**	3-Br-Ph	Et	NO_2
b	2-Br-Ph	Me	NO_2	**o**	4-Br-Ph	Et	MeO
c	3-Br-Ph	Me	MeO	**p**	4-Ph-Ph	Et	MeO
d	3-Br-Ph	Me	NO_2	**q**	3,5-Me_2-Ph	Et	MeO
e	4-Br-Ph	Me	MeO	**r**	3,5-Me_2-Ph	Et	NO_2
f	4-Ph-Ph	Me	MeO	**s**	3,5-Me_2-Ph	Et	H
g	3,5-Me_2-Ph	Me	MeO	**t**	3,5-$(CF_3)_2$-Ph	Et	MeO
h	3,5-Me_2-Ph	Me	NO_2	**u**	3,5-$(CF_3)_2$-Ph	Et	NO_2
i	3,5-$(CF_3)_2$-Ph	Me	MeO	**v**	2,6-Cl_2-Ph	Et	MeO
j	3,5-$(CF_3)_2$-Ph	Me	NO_2	**w**	2,6-Cl_2-Ph	Et	NO_2
k	2-Br-Ph	Et	MeO	**x**	1-naphthyl	Et	MeO
l	2-Br-Ph	Et	NO_2	**y**	1-naphthyl	*i*-Pr	MeO
m	3-Br-Ph	Et	MeO				

Scheme 1. Synthesis of compounds **5a**–**y**. Reagents and conditions: (a) (i) CDI, CH_3CN, rt, 30 min; (ii) $R_1CH(CO_2Et)(CO_2K)$, Et_3N, anhydrous $MgCl_2$, rt, overnight, then reflux, 2 h; (iii) 13% HCl; (b) (i) $R_1CHBrCOOEt$, Zn, THF, reflux, 8–16 h; (ii) 50% K_2CO_3; (iii) 13% HCl; (c) thiourea, EtONa, EtOH, reflux, 16 h. (d) (4-R_2)$PhCH_2X$ (X = Br or Cl), K_2CO_3, DMF, rt, 18 h.

activity. In particular, the 6-(3,5-dimethylbenzyl) analogue **5q** turned out to be the most potent with an IC_{50} value of 0.12 μM, being about 40-fold more active than the reference compounds HEPT and DDI. Based upon the preliminary SAR studies of these new 2-benzylsulfanyl-substituted *S*-DABOs, some structural requirements for high potency against HIV-1 were identified. These results provided useful indicators for guiding the further rational design of new *S*-DABO analogues as more active and selective HIV-1 inhibitors.

4. Experimental protocols

4.1. Chemistry

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. ^1H 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**–**e** and **6g** were obtained commercially; 3,5-dimethylpheny acetic acid (**6f**) was synthesized according to the literature procedure [11,12]; and β-ketoesters **7o** and **p** were prepared following the procedure described previously [9].

Table 1
Biological activities of compounds **5a**–**y** against HIV-1 and HIV-2 in MT-4 cells

Compound	Ar	R1	R2	IC_{50} (μM)[a]		CC_{50} (μM)[b]	SI[c]
				HIV-1 III_B	HIV-2		
5a	2-Br-Ph	Me	MeO	1.04 ± 0.02	>179	179 ± 7.35	172
5b	2-Br-Ph	Me	NO_2	1.46 ± 0.02	>246	≥246	≥168
5c	3-Br-Ph	Me	MeO	6.42 ± 0.39	>290	>290	>45
5d	3-Br-Ph	Me	NO_2	14.21 ± 9.93	>280	>280	>20
5e	4-Br-Ph	Me	MeO	>290	>290	>290	X1
5f	4-Ph-Ph	Me	MeO	>100	>100	100 ± 18	X1
5g	3,5-Me_2-Ph	Me	MeO	1.10 ± 0.24	>329	>329	>299
5h	3,5-Me_2-Ph	Me	NO_2	5.46 ± 2.48	>316	>316	>58
5i	3,5-$(CF_3)_2$-Ph	Me	MeO	>256	>256	>256	X1
5j	3,5-$(CF_3)_2$-Ph	Me	NO_2	>248	>248	>248	X1
5k	2-Br-Ph	Et	MeO	0.23 ± 0.02	>165 ± 26	165	717
5l	2-Br-Ph	Et	NO_2	0.74 ± 0.20	>272	>272	>368
5m	3-Br-Ph	Et	MeO	0.28 ± 0.02	>201	201 ± 21	718
5n	3-Br-Ph	Et	NO_2	1.34 ± 0.44	>187	187 ± 34	139.6
5o	4-Br-Ph	Et	MeO	>281	>281	>281	X1
5p	4-Ph-Ph	Et	MeO	>194	>194	194 ± 18	X1
5q	3,5-Me_2-Ph	Et	MeO	0.12 ± 0.03	>317	>317	>2642
5r	3,5-Me_2-Ph	Et	NO_2	0.16 ± 0.06	>305	>305	>1906
5s	3,5-Me_2-Ph	Et	H	0.95 ± 0.06	>343	>343	>361
5t	3,5-$(CF_3)_2$-Ph	Et	MeO	≥9.47	>249	>249	≤26
5u	3,5-$(CF_3)_2$-Ph	Et	NO_2	>242	>242	>242	X1
5v	2,6-Cl_2-Ph	Et	MeO	0.16 ± 0.08	>287	>287	>1793
5w	2,6-Cl_2-Ph	Et	NO_2	0.36 ± 0.17	>278	>278	>772
5x	1-Naphthyl	Et	MeO	1.32 ± 0.07	>227	261 ± 25	198
5y	1-Naphthyl	*i*-Pr	MeO	0.21 ± 0.004	>37.39	37.39 ± 4.08	178
HEPT				5.06 ± 0.19	>405.38	≤399	79
DDI				5.37 ± 0.1	2.71 ± 0.25	≥529	≥98

[a] IC_{50}: Inhibitory concentration of compound achieving 50% inhibition of HIV-1 multiplication in MT-4 infected cells.
[b] CC_{50}: Cytotoxic concentration of compound that reduces the normal uninfected MT-4 cell viability by 50%.
[c] SI: Selectivity index: ratio CC_{50}/IC_{50}.

*4.1.1. General procedure for the preparation of β-ketoesters **7a**–**n***

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 4–12 h 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), TEA (100 mL, 717 mmol) and $MgCl_2$ (54 g, 567 mmol) were added and the mixture was continued stirring at room temperature for 2 h. Then a solution of arylacetyl imidazolide, which was prepared from arylacetic acid (214 mmol) and 1,1′-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, 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**–**n** to be used directly in the next step without further purification.

*4.1.2. General procedure for the preparation of 5-alkyl-6-arylmethyl-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, 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.

*4.1.3. General procedure for the preparation of target compounds **5a**–**y***

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 benzyl halides (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**–**y**.

*4.1.3.1. 6-(2-Bromobenzyl)-2-(4-methoxybenzylsulfanyl)-5-methylpyrimidin-4(3H)-one (**5a**).* Yield 38%; recrystallized from AcOEt as a white solid; mp: 212.1–213.5 °C; FT-IR (KBr): *ν* 3421 (NH), 2950, 2928, 2834 (CH_3, CH_2), 1650 (C═O), 1610, 1551, 1512, 1474 (aryl) cm^{-1}; ^1H NMR ($CDCl_3$): *δ* (ppm) 1.98 (s, 3H, CH_3), 3.68 (s, 3H, OCH_3), 4.02 (s, 2H, $ArCH_2$), 4.07 (s, 2H, SCH_2), 6.69 (d, 2H, $J = 8.4$ Hz, $Ar'H_{3,5}$), 6.94 (d, 2H, $J = 8.4$ Hz, $Ar'H_{2,6}$), 7.16–7.64 (m, 4H, ArH), 12.53 (br s, 1H, CONH); ^{13}C NMR ($CDCl_3$): *δ* (ppm) 10.56 (CH_3), 33.88 (CH_2), 40.85 (CH_2), 55.15 (OCH_3), 113.78 (2C), 116.89 (C-5), 124.99, 127.22, 128.06, 128.95, 130.11 (2C), 130.98, 132.56, 137.59, 156.31 (C-6), 158.78 (Ar′-C_4), 160.84 (C-2), 165.42 (C-4); MS (EI): *m/z* 430 (M^+); Anal. Calcd for $C_{20}H_{19}BrN_2O_2S$ (430.04): C, 55.69; H, 4.44; N, 6.49; S, 7.43. Found: C, 55.65; H, 4.47; N, 6.46; S, 7.45%.

*4.1.3.2. 6-(2-Bromobenzyl)-5-methyl-2-(4-nitrobenzylsulfanyl)-pyrimidin-4(3H)-one (**5b**).* Yield 77%; recrystallized from MEK as a yellow needle crystal; mp: 218.3–218.9 °C; FT-IR (KBr): *ν* 3431 (NH), 2932, 2848 (CH_2), 1661 (C═O), 1598, 1578, 1511, 1514 (aryl)

cm^{-1}; ^{1}H NMR (DMSO-d_6): δ (ppm) 1.99 (s, 3H, CH_3), 4.02 (s, 2H, ArCH_2), 4.26 (s, 2H, SCH_2), 7.14–7.61 (m, 6H, Ar′$H_{2,6}$, ArH), 7.97 (d, 2H, Ar′$H_{3,5}$), 12.66 (br s, 1H CONH); ^{13}C NMR (DMSO-d_6): δ (ppm) 10.47 (*C*H$_3$), 32.20 (*C*H$_2$), 115.83 (C-5), 123.32 (2C), 124.63, 127.72, 128.60, 129.88 (2C), 131.78, 132.47, 137.84, 146.45, 146.62, 156.2 (C-6), 159.53 (C-2), 163.50 (C-4); MS (EI): m/z 445 (M$^+$); Anal. Calcd for $C_{19}H_{16}BrN_3O_3S$ (445.01): C, 51.13; H, 3.61; N, 9.41; S, 7.18. Found: C. 51.15; H, 3.62; N, 9.38; S, 7.16%.

*4.1.3.3. 6-(3-Bromobenzyl)-2-(4-methoxybenzylsulfanyl)-5-methylpyrimidin-4(3*H*)-one (**5c**).* Yield 53%; recrystallized from AcOEt as a white needle crystal; mp: 176.2–176.3 °C; FT-IR (KBr): ν 3408 (NH), 2946, 2927, 2832 (CH$_3$, CH$_2$), 1655 (C═O), 1610, 1553, 1510 (aryl) cm^{-1}; ^{1}H NMR (DMSO-d_6) δ (ppm) 1.96 (s, 3H, CH_3), 3.71 (s, 3H, OCH_3), 3.92 (s, 2H, ArCH_2), 4.22 (s, 2H, CH_2), 6.75 (d, 2H, J = 8.0 Hz, Ar′$H_{3,5}$), 7.11 (d, 2H, J = 8.0 Hz, Ar′$H_{2,6}$), 7.26 (m, 2H, Ar$H_{5,6}$), 7.43–7.45 (m, 1H, ArH_4), 7.49 (s, 1H, ArH_2), 12.58 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6): δ (ppm) 10.57 (*C*H$_3$), 32.97 (*C*H$_2$), 55.13 (OCH$_3$), 113.85 (2C), 115.62 (C-5), 121.79, 128.25, 129.33, 129.72, 130.17 (2C), 130.57, 131.82, 141.19, 157.00 (C-6), 158.50 (Ar′-C$_4$), 160.00 (C-2), 163.47 (C-4); MS (EI): m/z 430 (M$^+$); Anal. Calcd for $C_{20}H_{19}BrN_2O_2S$ (430.04): C, 55.69; H, 4.44; N, 6.49; S, 7.43. Found: C, 55.72; H, 4.43; N, 6.46; S, 7.48%.

*4.1.3.4. 6-(3-Bromobenzyl)-5-methyl-2-(4-nitrobenzylsulfanyl)-pyrimidin-4(3*H*)-one (**5d**).* Yield 54%; recrystallized from MEK as a white solid; mp: 207.6–209.0 °C; FT-IR (KBr): ν 3432 (NH), 2938, 2851 (CH$_2$), 1656 (C═O), 1595, 1545, 1518, 1476 (aryl) cm^{-1}; ^{1}H NMR (DMSO-d_6): δ (ppm) 1.95 (s, 3H, CH_3), 3.91 (s, 2H, ArCH_2), 4.39 (s, 2H, SCH_2), 7.19–7.42 (m, 4H, ArH), 7.46 (d, 2H, J = 8.4 Hz, Ar′$H_{2,6}$), 8.03 (d, 2H, J = 8.4 Hz, Ar′$H_{3,5}$), 12.69 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6): δ (ppm) 10.51 (*C*H$_3$), 32.54 (*C*H$_2$), 115.72 (C-5), 121.75, 123.40 (2C), 128.23, 129.30, 130.05 (2C), 130.53, 131.70, 141.13, 146.50, 146.58, 156.46 (C-6), 160.07 (C-2), 163.53 (C-4); MS (EI): m/z 445 (M$^+$); Anal. Calcd for $C_{19}H_{16}BrN_3O_3S$ (445.01): C, 51.13; H, 3.61; N, 9.41; S, 7.18. Found: C, 51.10; H, 3.63; N, 9.45; S, 7.21%.

*4.1.3.5. 6-(4-Bromobenzyl)-2-(4-methoxybenzylsulfanyl)-5-methylpyrimidin-4(3*H*)-one (**5e**).* Yield 66%; recrystallized from AcOEt as a white solid; mp: 204.1–205.6 °C; FT-IR: (KBr) ν 3433 (NH), 2932, 2837 (CH$_2$), 1653 (C═O), 1608, 1549, 1510, 1486 (aryl); ^{1}H NMR (DMSO-d_6): δ (ppm) 1.97 (s, 3H, CH_3), 3.72 (s, 3H, OCH_3), 3.87 (s, 2H, ArCH_2), 4.24 (s, 2H, SCH_2), 6.74 (d, 2H, J = 8.4 Hz, Ar′$H_{3,5}$), 7.10 (d, 2H, J = 8.4 Hz, Ar′$H_{2,6}$), 7.20 (d, 2H, J = 8.0 Hz, Ar$H_{2,6}$), 7.47 (d, 2H, J = 8.0 Hz, Ar$H_{3,5}$), 12.55 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6): δ (ppm) 10.57 (*C*H$_3$), 32.86 (*C*H$_2$), 55.15 (OCH$_3$), 113.79 (2C), 115.50 (C-5), 119.55, 129.77, 130.15 (2C), 131.30 (2C), 131.35 (2C), 137.92, 156.79 (C-6), 158.46 (Ar′-C$_4$), 160.17 (C-2), 163.43 (C-4); MS (EI): m/z 430 (M$^+$); Anal. Calcd for $C_{20}H_{19}BrN_2O_2S$ (430.04): C, 55.69; H, 4.44; N, 6.49; S, 7.43. Found: C, 55.67; H, 4.46; N, 6.49; S, 7.41%.

*4.1.3.6. 2-(4-Methoxybenzylsulfanyl)-5-methyl-6-(4-phenylbenzyl)-pyrimidin-4(3*H*)-one (**5f**).* Yield 50%; recrystallized from CHCl$_3$ as a white solid; mp: 215.2–216.9 °C; FT-IR (KBr): ν 3421 (NH), 2954, 2928, 2834 (CH$_3$, CH$_2$), 1659 (C═O), 1582, 1542, 1509, 1486 (aryl) cm^{-1}; ^{1}H NMR (DMSO-d_6): δ (ppm) 2.00 (s, 3H, CH_3), 3.61 (s, 3H, OCH_3), 3.94 (s, 2H, ArCH_2), 4.27 (s, 2H, SCH_2), 6.69 (d, 2H, J = 8.0 Hz, Ar′$H_{3,5}$), 7.13 (d, 2H, J = 8.0 Hz, Ar′$H_{2,6}$), 7.36–7.65 (m, 9H, ArH), 12.56 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6): δ (ppm) 10.69 (*C*H$_3$), 32.90 (*C*H$_2$), 55.07 (OCH$_3$), 113.84 (2C), 115.84 (C-5), 126.65 (2C), 126.85 (2C), 127.46, 129.08 (2C), 129.68 (2C), 129.89, 130.23 (2C), 137.78, 138.33, 140.09, 156.09 (C-6), 158.46 (Ar′-C$_4$), 160.45 (C-2), 163.37 (C-4); MS (EI): m/z 428 (M$^+$); Anal. Calcd for $C_{26}H_{24}N_2O_2S$ (428.16): C, 72.87; H, 5.64; N, 6.54; S, 7.48. Found: C, 72.91; H, 5.61; N, 6.53; S, 7.45%.

*4.1.3.7. 6-(3,5-Dimethylbenzyl)-2-(4-methoxybenzylsulfanyl)-5-methylpyrimidin-4(3*H*)-one (**5g**).* Yield 44%; column chromatography (eluent CH$_2$Cl$_2$/AcOEt 100/10), off-white solid; mp: 167.0–167.4 °C; FT-IR (KBr): ν 3422 (NH), 2942, 2920, 2832 (CH$_3$, CH$_2$), 1653 (C═O), 1609, 1570, 1549, 1510, 1453 (aryl) cm^{-1}; ^{1}H NMR (CDCl$_3$): δ (ppm) 1.96 (s, 3H, CH_3), 2.22 (s, 6H, Ar(CH_3)$_2$), 3.71 (s, 3H, OCH_3), 3.82 (s, 2H, ArCH_2), 4.27 (s, 2H, SCH_2), 6.75 (d, 2H, J = 8.8 Hz, Ar′$H_{3,5}$), 6.86 (s, 3H, Ar$H_{2,4,6}$), 7.17 (d, 2H, J = 8.8 Hz, Ar′$H_{2,6}$), 12.51 (br s, 1H, CONH); ^{13}C NMR (CDCl$_3$): δ (ppm) 10.75 (*C*H$_3$), 21.29 (2C, Ar(*C*H$_3$)$_2$), 34.20 (*C*H$_2$), 40.92 (*C*H$_2$), 55.22 (OCH$_3$), 113.92 (2C), 116.59 (C-5), 126.76 (2C), 128.11, 129.03, 130.25 (2C), 137.76, 137.96 (2C), 155.76 (C-6), 158.95 (Ar′-C$_4$), 162.15 (C-2), 164.99 (C-4); MS (EI): m/z 380 (M$^+$); Anal. Calcd for $C_{22}H_{24}N_2O_2S$ (380.16): C, 69.44; H, 6.36; N, 7.36; S, 8.43. Found: C, 69.46; H, 6.33; N, 7.40; S, 8.42%.

*4.1.3.8. 6-(3,5-Dimethylbenzyl)-5-methyl-2-(4-nitrobenzylsulfanyl)-pyrimidin-4(3*H*)-one (**5h**).* Yield 47.41%; column chromatography (eluent CH$_2$Cl$_2$/AcOEt 100/10), off-white solid; mp: 225.6–226.4 °C; FT-IR (KBr): ν 3408 (NH), 2921, 2852 (CH$_2$), 1655 (C═O), 1599, 1544, 1510 (aryl) cm^{-1}; ^{1}H NMR (CDCl$_3$): δ (ppm) 1.95 (s, 3H, CH_3), 2.18 (s, 6H, Ar(CH_3)$_2$), 3.81 (s, 2H, ArCH_2), 4.42 (s, 2H, SCH_2), 6.78 (s, 2H, Ar$H_{2,6}$), 6.83 (s, 1H, ArH_4), 7.49 (d, 2H, J = 8.0 Hz, Ar′$H_{2,6}$), 8.00 (d, 2H, J = 8.0 Hz, Ar′$H_{3,5}$), 12.58 (br s, 1H, CONH); ^{13}C NMR (CDCl$_3$): δ (ppm) 10.78 (*C*H$_3$), 21.25 (2C, Ar(*C*H$_3$)$_2$), 33.50 (*C*H$_2$), 40.69 (*C*H$_2$), 117.24 (C-5), 123.56 (2C), 126.69 (2C), 128.31, 129.84 (2C), 137.54, 138.15 (2C), 145.03, 147.10, 154.32 (C-6), 161.97 (C-2), 164.55 (C-4); MS (EI): m/z 395 (M$^+$); Anal. Calcd for $C_{21}H_{21}N_3O_3S$ (395.13): C, 63.78; H, 5.35; N, 10.63; S, 8.11. Found: C, 63.75; H, 5.38; N, 10.61; S, 8.15%.

*4.1.3.9. 6-(3,5-Bis(trifluoromethyl)benzyl)-2-(4-methoxybenzylsulfanyl)-5-methylpyrimidin-4(3*H*)-one (**5i**).* Yield 31%; recrystallized form AcOEt as a white solid; mp: 222.2–222.6 °C; FT-IR (KBr): ν 3422 (NH), 2965, 2938, 2837 (CH$_3$, CH$_2$), 1656 (C═O), 1578, 1557, 1514, 1465 (aryl) cm^{-1}; ^{1}H NMR (DMSO-d_6): δ (ppm) 2.0 (s, 3H, CH_3), 3.67 (s, OCH_3), 4.10 (s, 2H, ArCH_2), 4.15 (s, 2H, SCH_2), 6.68 (d, 2H, J = 8.4 Hz, Ar′$H_{3,5}$), 6.95 (d, 2H, J = 8.4 Hz, Ar′$H_{2,6}$), 7.92 (s, 1H, ArH_4), 8.02 (s, 2H, Ar$H_{2,6}$), 12.58 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6): δ (ppm) 10.39 (*C*H$_3$), 32.85 (*C*H$_2$), 38.89 (*C*H$_2$), 55.04 (OCH$_3$), 113.74 (2C, Ar′-C$_{3,5}$), 115.65 (C-5), 120.16 (sept, J = 3.7 Hz, Ar-C$_4$), 123.55 (q, 2C, J = 271.1 Hz, *C*F$_3$), 129.35, 129.84 (2C, Ar′-C$_{2,6}$), 130.14 (q, 2C, J = 32.2 Hz, Ar-C$_{3,5}$), 130.58 (2C, Ar-C$_{2,6}$), 141.89, 157.18 (C-6), 158.51 (Ar′-C$_4$), 158.82 (C-2), 163.27 (C-4); MS (EI): m/z 488 (M$^+$); Anal. Calcd for $C_{22}H_{18}F_6N_2O_2S$ (488.1): C, 54.10; H, 3.71; N, 5.74; S, 6.56. Found: C, 54.15; H, 3.68; N, 5.77; S, 6.51%.

*4.1.3.10. 6-(3,5-Bis(trifluoromethyl)benzyl)-5-methyl-2-(4-nitrobenzylsulfanyl)pyrimidin-4(3*H*)-one (**5j**).* Yield 21%; recrystallized from AcOEt as a white needle crystal; mp: 234.4–235.2 °C; FT-IR (KBr): ν 3421 (NH), 2985, 2928, 2850 (CH$_3$, CH$_2$), 1648 (C═O), 1597, 1549, 1517 (aryl) cm^{-1}; ^{1}H NMR (DMSO-d_6): δ (ppm) 2.00 (s, 3H, CH_3), 4.17 (s, 2H, ArCH_2), 4.30 (s, 2H, SCH_2), 7.31 (d, 2H, J = 8.4 Hz, Ar′$H_{2,6}$), 7.87 (s,1H, ArH_4), 7.94 (s, 2H, Ar$H_{2,6}$), 7.96 (d, 2H, J = 8.4 Hz, Ar′$H_{3,5}$), 12.75 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6): δ (ppm) 10.36 (*C*H$_3$), 32.32 (*C*H$_2$), 38.88 (*C*H$_2$), 115.93 (C-5), 120.07 (sept, J = 3.6 Hz, Ar-C$_4$), 123.21 (2C), 123.46 (q, 2C, J = 271.1 Hz, *C*F$_3$), 129.69 (2C), 130.08 (q, 2C, J = 32.36 Hz, Ar-C$_{3,5}$), 130.39 (2C, Ar-C$_{2,6}$), 141.86, 146.27, 146.44, 156.75 (C-6), 159.07 (C-2), 163.43 (C-4); MS (EI): m/z 503 (M$^+$); Anal. Calcd for $C_{21}H_{15}F_6N_3O_3S$ (503.07): C, 50.10; H, 3.00; N, 8.35; S, 6.37. Found: C, 50.06; H, 3.03; N, 8.35; S, 6.33%.

*4.1.3.11. 6-(2-Bromobenzyl)-5-ethyl-2-(4-methoxybenzylsulfanyl)-pyrimidin-4(3*H*)-one (**5k**).* Yield 73%; column chromatography (eluent CH$_2$Cl$_2$/AcOEt 100/10), white solid; mp: 149.7–149.8 °C; FT-IR (KBr): ν 3397 (NH), 2965, 2933, 2870, 2836 (CH$_3$, CH$_2$), 1650 (C═O), 1612, 1567, 1512 (aryl) cm^{-1}; ^{1}H NMR (CDCl$_3$): δ (ppm) 1.10

(t, 3H, $J = 7.6$ Hz, CH_2CH_3), 2.56 (q, 2H, $J = 7.6$ Hz, CH_2CH_3), 3.76 (s, 3H, OCH_3), 4.08 (s, 2H, $ArCH_2$), 4.16 (s, 2H, SCH_2), 6.71 (d, 2H, $J = 8.4$ Hz, Ar′$H_{3,5}$), 7.02 (d, 2H, $J = 8.4$ Hz, Ar′$H_{2,6}$), 7.06–7.58 (m, 4H, Ar*H*), 12.70 (br s, 1H, CON*H*); ^{13}C NMR ($CDCl_3$): δ (ppm) 12.98 (CH_2CH_3), 18.67 (CH_2CH_3), 33.85 (CH_2), 40.25 (CH_2), 55.18 (OCH_3), 113.79 (2C), 122.84 (C-5), 125.07, 127.22, 128.04, 129.06, 130.13 (2C), 131.00, 132.58, 137.94, 156.45 (C-4), 158.78 (Ar′-C_4), 160.41 (C-2), 165.02 (C-4); MS (EI): m/z 444 (M^+); Anal. Calcd for $C_{21}H_{21}BrN_2O_2S$ (444.05): C, 56.63; H, 4.75; N, 6.29; S, 7.20. Found: C, 56.66; H, 4.74; N, 6.27; S, 7.23%.

*4.1.3.12. 6-(2-Bromobenzyl)-5-ethyl-2-(4-nitrobenzylsulfanyl)pyrimidin-4(3*H*)-one (**5l**).* Yield 63%; recrystallized from AcOEt as white solid; mp: 187.6–188.8 °C; FT-IR (KBr): ν 3421 (NH), 2964, 2929, 2867 (CH_2, CH_2), 1638 (C═O), 1579, 1545, 1521, 1458 (aryl) cm^{-1}; ^{1}H NMR ($CDCl_3$): δ (ppm) 1.13 (t, 3H, $J = 7.4$ Hz, CH_2CH_3), 2.59 (q, 2H, $J = 7.4$ Hz, CH_2CH_3), 4.08 (s, 2H, $ArCH_2$), 4.23 (s, 2H, SCH_2), 7.02–7.58 (m, 6H, Ar′$H_{2,6}$, Ar*H*), 7.95 (d, 2H, $J = 8.8$ Hz, Ar′$H_{3,5}$), 12.44 (br s, 1H, CON*H*). ^{13}C NMR ($CDCl_3$): δ (ppm) 12.97 (CH_2CH_3), 18.71 (CH_2CH_3), 33.21 (CH_2), 40.16 (CH_2), 123.34 (C-5), 123.51 (2C), 125.24, 127.30, 128.30, 129.64 (2C), 130.96, 132.75, 137.76, 145.14, 146.98, 155.13 (C-6), 160.34 (C-2), 164.74 (C-4). MS (EI): m/z 459 (M^+); Anal. Calcd for $C_{20}H_{18}BrN_3O_3S$ (459.03): C, 52.18; H, 3.94; N, 9.13; S, 6.97. Found: C, 52.16; H, 3.95; N, 9.15; S, 6.93%.

*4.1.3.13. 6-(3-Bromobenzyl)-5-ethyl-2-(4-methoxybenzylsulfanyl)-pyrimidin-4(3*H*)-one (**5m**).* Yield 68%; column chromatography (eluent petroleum ether/acetone 100/10), white solid; mp: 151.5–152.5 °C; FT-IR (KBr): ν 3412 (NH), 2959, 2933, 2867, 2836 (CH_3, CH_2), 1650 (C═O), 1611, 1582, 1552, 1512, 1475 (aryl) cm^{-1}; ^{1}H NMR (DMSO-d_6): δ (ppm) 1.08 (t, 3H, $J = 7.2$ Hz, CH_2CH_3), 2.55 (q, 2H, $J = 7.2$ Hz, CH_2CH_3), 3.38 (s, 3H, OCH_3), 3.90 (s, 2H, $ArCH_2$), 4.28 (s, 2H, SCH_2), 6.75 (d, 2H, $J = 8.4$ Hz, Ar′$H_{3,5}$), 7.11 (d, 2H, $J = 8.4$ Hz, Ar′$H_{2,6}$), 7.16 (m, 2H, Ar$H_{5,6}$), 7.37 (m, 1H, ArH_4), 7.43 (s, 1H, ArH_2), 12.54 (br s, 1H, CON*H*); ^{13}C NMR (DMSO-d_6): δ (ppm) 13.32 (CH_2CH_3), 18.29 (CH_2CH_3), 32.91 (CH_2), 55.14 (OCH_3), 113.86 (2C), 121.58 (C-5), 121.74, 128.29, 129.31, 129.69, 130.16 (2C), 130.57, 131.88, 141.50, 157.16 (C-6), 158.49 (Ar′-C_4), 159.48 (C-2), 163.01 (C-4); MS (EI): m/z 444 (M^+); Anal. Calcd for $C_{21}H_{21}BrN_2O_2S$ (444.05): C, 56.63; H, 4.75; N, 6.29; S, 7.20. Found: C, 56.65; H, 4.77; N, 6.25; S, 7.23%.

*4.1.3.14. 6-(3-Bromobenzyl)-5-ethyl-2-(4-nitrobenzylsulfanyl)pyrimidin-4(3*H*)-one (**5n**).* Yield 57%; recrystallized from CH_3CN as an off-white needle crystal; mp: 162.6–163.8 °C; FT-IR (KBr): ν 3417 (NH), 2964, 2933, 2850 (CH_3, CH_2), 1660 (C═O), 1597, 1551, 1517, 1472 (aryl) cm^{-1}; ^{1}H NMR (DMSO-d_6): δ (ppm) 0.94 (t, 3H, $J = 7.6$ Hz, CH_2CH_3), 2.44 (q, 2H, $J = 7.6$ Hz, CH_2CH_3), 3.92 (s, 2H, $ArCH_2$), 4.38 (s, 2H, SCH_2), 7.19–7.41 (m, 4H, Ar*H*), 7.44 (d, 2H, $J = 8.4$ Hz, Ar′$H_{2,6}$), 8.03 (d, 2H, $J = 8.4$ Hz, Ar′$H_{3,5}$), 12.65 (br s, 1H, CON*H*); ^{13}C NMR (DMSO-d_6): δ (ppm) 13.27 (CH_2CH_3), 18.26 (CH_2CH_3), 32.49 (CH_2), 38.99 (CH_2), 121.74, 122.17 (C-5), 123.45 (2C), 128.27, 129.30, 130.07 (2C), 130.57, 131.77, 141.51, 146.51, 146.60, 155.78 (C-6), 159.09 (C-2), 162.63 (C-4); MS (EI): m/z 459 (M^+); Anal. Calcd for $C_{20}H_{18}BrN_3O_3S$ (459.03): C, 52.18; H, 3.94; N, 9.13; S, 6.97. Found: C, 52.15; H, 3.96; N, 9.15; S, 6.95%.

*4.1.3.15. 6-(4-Bromobenzyl)-5-ethyl-2-(4-methoxybenzylsulfanyl)-pyrimidin-4(3*H*)-one (**5o**).* Yield 52%; recrystallized from AcOEt as a white solid; mp: 187.4–188.0 °C; FT-IR (KBr): ν 3433 (NH), 2970, 2931, 2867, 2835 (CH_3, CH_2), 1660 (C═O), 1609, 1579, 1545, 1511, 1484 (aryl) cm^{-1}; ^{1}H NMR (DMSO-d_6): δ (ppm) 0.96 (t, 3H, $J = 7.4$ Hz, CH_2CH_3), 2.46 (q, 2H, $J = 7.4$ Hz, CH_2CH_3), 3.72 (s, 3H, OCH_3), 3.88 (s, 2H, $ArCH_2$), 4.22 (s, 2H, SCH_2), 6.74 (d, 2H, $J = 8.4$ Hz, Ar′$H_{3,5}$), 7.07 (d, 2H, $J = 8.4$ Hz, Ar′$H_{2,6}$), 7.21 (d, 2H, $J = 8.4$ Hz, Ar$H_{2,6}$), 7.48 (d, 2H, $J = 8.4$ Hz, Ar$H_{3,5}$), 12.53 (br s, 1H, CON*H*); ^{13}C NMR (DMSO-d_6): δ (ppm) 13.40 (CH_2CH_3), 18.31 (CH_2CH_3), 32.81 (CH_2), 38.94 (CH_2), 55.15 (OCH_3), 113.81(2C), 119.56, 121.54 (C-5), 129.78, 130.14 (2C), 131.34 (4C), 138.21, 157.02 (C-6), 158.47 (Ar′-C_4), 159.96 (C-2), 163.03 (C-4); MS (EI): m/z 444 (M^+); Anal. Calcd for $C_{21}H_{21}BrN_2O_2S$ (444.05): C, 56.63; H, 4.75; N, 6.29; S, 7.20. Found: C, 56.67; H, 4.73; N, 6.27; S, 7.21%.

*4.1.3.16. 5-Ethyl-2-(4-methoxybenzylsulfanyl)-6-(4-phenylbenzyl)-pyrimidin-4(3*H*)-one (**5p**).* Yield 18%; recrystallized from THF as a white solid; mp: 222.0–223.2 °C; FT-IR (KBr): ν 3398 (NH), 2973, 2932, 2867 (CH_3, CH_2), 1654 (C═O), 1609, 1583, 1542, 1509, 1486 (aryl) cm^{-1}; ^{1}H NMR (DMSO-d_6): δ (ppm) 1.00 (t, 3H, $J = 7.4$ Hz, CH_2CH_3), 2.50 (q, 2H, $J = 7.4$ Hz, CH_2CH_3), 3.60 (s, 3H, OCH_3), 3.95 (s, 2H, $ArCH_2$), 4.26 (s, 2H, SCH_2), 6.68 (d, 2H, $J = 8.8$ Hz, Ar′$H_{3,5}$), 7.10 (d, 2H, $J = 8.8$ Hz, Ar′$H_{2,6}$), 7.34–7.66 (m, 9H, Ar*H*), 12.53 (br s, 1H, CON*H*); ^{13}C NMR (DMSO-d_6): δ (ppm) 13.43 (CH_2CH_3), 18.34 (CH_2CH_3), 32.78 (CH_2), 55.00 (OCH_3), 113.80 (2C), 122.11 (C-5), 126.59 (2C), 126.75 (2C), 127.40, 129.02 (2C), 129.68 (2C), 129.84, 130.16 (2C), 138.03, 138.23, 140.01, 156.14 (C-6), 158.40 (Ar′-C_4), 159.53 (C-2), 162.82 (C-4); MS (EI): m/z 442 (M^+); Anal. Calcd for $C_{27}H_{26}N_2O_2S$ (442.17): C, 73.27; H, 5.92; N, 6.33; S, 7.25. Found: C, 73.30; H, 5.91; N, 6.31; S, 7.25%.

*4.1.3.17. 6-(3,5-Dimethylbenzyl)-5-ethyl-2-(4-methoxybenzylsulfanyl)-pyrimidin-4(3*H*)-one (**5q**).* Yield 57%; column chromatography (eluent CH_2Cl_2/AcOEt 100/10), white solid; mp: 146.3–147.4 °C; FT-IR (KBr): ν 3422 (NH), 2969, 2932, 2874, 2832 (CH_3, CH_2), 1647 (C═O), 1613, 1578, 1542, 1511, 1458 (aryl) cm^{-1}; ^{1}H NMR ($CDCl_3$): δ (ppm) 1.02 (t, 3H, $J = 7.6$ Hz, CH_2CH_3), 2.27(s, 6H, Ar(CH_3)$_2$), 2.51(q, 2H, $J = 7.6$ Hz, CH_2CH_3), 3.76 (s, 3H, OCH_3), 3.88 (s, 2H, $ArCH_2$), 4.32 (s, 2H, SCH_2), 6.79 (d, 2H, $J = 8.4$ Hz, Ar′$H_{3,5}$), 6.93 (s, 3H, Ar$H_{2,4,6}$), 7.20 (d, 2H, $J = 8.4$ Hz, Ar′$H_{2,6}$), 12.60 (1H, br s, CON*H*); ^{13}C NMR ($CDCl_3$): δ (ppm) 13.18 (CH_2CH_3), 18.81 (CH_2CH_3), 21.28 (2C, Ar(CH_3)$_2$), 34.09 (CH_2), 40.31 (CH_2), 55.20 (OCH_3), 113.90 (2C), 122.51 (C-5), 126.87 (2C), 128.05, 129.14, 130.24 (2C), 137.89 (2C), 138.12, 155.92 (C-6), 158.91 (Ar′-C_4), 161.62 (C-2), 164.70 (C-4); MS (EI): m/z 394 (M^+); Anal. Calcd for $C_{23}H_{26}N_2O_2S$ (394.17): C, 70.02; H, 6.64; N, 7.10; S, 8.13. Found: C, 70.08; H, 6.61; N, 7.13; S, 8.15%.

*4.1.3.18. 6-(3,5-Dimethylbenzyl)-5-ethyl-2-(4-nitrobenzylsulfanyl)-pyrimidin-4(3*H*)-one (**5r**).* Yield 48%; column chromatography (eluent CH_2Cl_2/AcOEt 100/10), off-white solid; mp: 193.7–194.1 °C; FT-IR (KBr): ν 3432 (NH), 2976, 2935, 2840 (CH_3, CH_2), 1653 (C═O), 1597, 1541, 1510 (aryl) cm^{-1}; ^{1}H NMR ($CDCl_3$): δ (ppm) 1.10 (t, 3H, $J = 7.4$ Hz, CH_2CH_3), 2.25 (s, 6H, Ar(CH_3)$_2$), 2.58 (q, 2H, $J = 7.4$ Hz, CH_2CH_3), 3.86 (s, 2H, $ArCH_2$), 4.34 (s, 2H, SCH_2), 6.77 (s, 2H, Ar$H_{2,6}$), 6.89 (s, 1H, ArH_4), 7.27 (d, 2H, $J = 8.8$ Hz, Ar′$H_{2,6}$), 7.95 (d, 2H, $J = 8.8$ Hz, Ar′$H_{3,5}$), 12.42 (br s, 1H, CON*H*); ^{13}C NMR ($CDCl_3$): δ (ppm) 13.20 (CH_2CH_3), 18.79 (CH_2CH_3), 21.24 (2C, Ar(CH_3)$_2$), 33.33 (CH_2), 40.11 (CH_2), 122.99 (C-5), 123.51 (2C), 126.78 (2C), 128.24, 129.82 (2C), 137.90, 138.09 (2C), 145.21, 147.03, 154.78 (C-6), 161.69 (C-2), 164.90 (C-4); MS (EI): m/z 409 (M^+); Anal. Calcd for $C_{22}H_{23}N_3O_3S$ (409.15): C, 64.53; H, 5.66; N, 10.26; S, 7.83. Found: C, 64.55; H, 5.69; N, 10.21; S, 7.85%.

*4.1.3.19. 2-(Benzylsulfanyl)-6-(3,5-dimethylbenzyl)-5-ethylpyrimidin-4(3*H*)-one (**5s**).* Yield 31%; column chromatography (eluent CH_2Cl_2/AcOEt 100/10), off-white solid; mp: 141.8–142.5 °C; FT-IR (KBr): ν 3423 (NH), 2966, 2929, 2843 (CH_3, CH_2), 1632 (C═O), 1571, 1550, 1493, 1454 (aryl) cm^{-1}; ^{1}H NMR ($CDCl_3$): δ (ppm) 1.12 (t, 3H, $J = 7.2$ Hz, CH_2CH_3), 2.31 (s, 6H, Ar(CH_3)$_2$), 2.62 (q, 2H, $J = 7.2$ Hz, CH_2CH_3), 3.91 (s, 2H, $ArCH_2$), 4.40 (s, 2H, SCH_2), 6.91 (s, 3H, Ar$H_{2,4,6}$), 7.27 (m, 5H, Ar′*H*), 12.92 (br s, 1H, CON*H*); ^{13}C NMR ($CDCl_3$): δ (ppm) 13.16 (CH_2CH_3), 18.75 (CH_2CH_3), 21.26 (2C, Ar(CH_3)$_2$), 34.33 (CH_2), 40.30 (CH_2), 122.36 (C-5), 126.80 (2C), 127.27, 128.02, 128.41 (2C), 129.05 (2C), 137.23, 137.84 (2C), 138.07, 155.99 (C-6), 161.80 (C-2),

165.27 (C-4); MS (EI): *m/z* 364 (M^+); Anal. Calcd for $C_{22}H_{24}N_2OS$ (364.16): C, 72.49; H, 6.64; N, 7.69; S, 8.80. Found: C, 72.48; H, 6.67; N, 7.63; S, 8.83%.

*4.1.3.20. 6-(3,5-Bis(trifluoromethyl)benzyl)-5-ethyl-2-(4-methoxybenzylsulfanyl)pyramidin-4(3H)-one (**5t**).* Yield 43%; column chromatography (eluent petroleum ether/acetone 100/10); mp: 199.7–201.4 °C; FT-IR (KBr): *ν* 3418 (NH), 2981, 2938, 2837 (CH_3, CH_2), 1654 (C=O), 1613, 1553, 1514, 1466 (aryl) cm^{-1}; ^1H NMR ($CDCl_3$): *δ* (ppm) 1.11 (t, 3H, $J = 7.2$ Hz, CH_2CH_3), 2.59 (q, 2H, $J = 7.2$ Hz, CH_2CH_3), 3.77 (s, 3H, OCH_3), 4.04 (s, 2H, $ArCH_2$), 4.22 (s, 2H, SCH_2), 6.75 (d, 2H, $J = 8.8$ Hz, $Ar'H_{3,5}$), 7.07 (d, 2H, $J = 8.8$ Hz, $Ar'H_{2,6}$), 7.76 (s, 3H, $ArH_{2,4,6}$), 12.70 (br s, 1H, CONH); ^{13}C NMR ($CDCl_3$): *δ* (ppm) 13.23 (CH_2CH_3), 18.76 (CH_2CH_3), 34.34 (CH_2), 39.64 (CH_2), 55.20 (OCH_3), 114.00 (2C, Ar′-$C_{3,5}$), 120.61 (sept, $J = 3.6$ Hz, Ar-C_4), 122.87 (C-5), 123.32 (q, 2C, $J = 270.8$ Hz, CF_3), 127.94, 129.53 (2C, Ar-$C_{2,6}$), 129.97 (2C, Ar′-$C_{2,6}$), 131.63 (q, 2C, $J = 33.37$ Hz, Ar-$C_{3,5}$), 140.61, 157.23 (C-6), 159.11 (Ar′-C_4), 159.20 (C-2), 164.56 (C-4); MS (EI): *m/z* 502 (M^+); Anal. Calcd for $C_{23}H_{20}F_6N_2O_2S$ (502.11): C, 54.98; H, 4.01; N, 5.58; S, 6.38. Found: C, 54.95; H, 4.02; N, 5.55; S, 6.36%.

*4.1.3.21. 6-(3,5-Bis(trifluoromethyl)benzyl)-5-ethyl-2-(4-nitrobenzylsulfanyl)pyrimidin-4(3H)-one (**5u**).* Yield 32%; recrystallized from AcOEt as an off-white solid; mp: 213–214.5 °C; FT-IR (KBr): *ν* 3408 (NH), 2976, 2935, 2874, 2855 (CH_3, CH_2), 1647 (C=O), 1599, 1581, 1543, 1518, 1460 (aryl) cm^{-1}; ^1H NMR (DMSO-d_6): *δ* (ppm) 0.98 (t, 3H, CH_2CH_3), 2.50 (m, 2H, CH_2CH_3), 4.18 (s, 2H, $ArCH_2$), 4.30 (s, 2H, SCH_2), 7.33 (d, 2H, $J = 8.8$ Hz, $Ar'H_{2,6}$), 7.87 (s, 1H, ArH_4), 7.93 (s, 2H, $ArH_{2,6}$), 7.96 (d, 2H, $J = 8.8$ Hz, $Ar'H_{3,5}$), 12.71 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6): *δ* (ppm) 13.11 (CH_2CH_3), 18.09 (CH_2CH_3), 32.34 (CH_2), 38.34 (CH_2), 120.08 (Ar-C_4), 122.38 (C-5), 123.23 (2C), 123.46 (q, 2C, $J = 271$ Hz, CF_3), 129.72 (2C), 130.05 (q, 2C, $J = 32.4$ Hz, Ar-$C_{3,5}$), 130.36 (2C, Ar-$C_{2,6}$), 142.10, 146.21, 146.44, 156.13 (C-6), 157.89 (C-2), 162.44 (C-4); MS (EI): *m/z* 517 (M^+); Anal. Calcd for $C_{22}H_{17}F_6N_3O_3S$ (517.09): C, 51.07; H, 3.31; N, 8.12; S, 6.20. Found: C, 51.06; H, 3.32; N, 8.15; S, 6.22%.

*4.1.3.22. 6-(2,6-Dichlorobenzyl)-5-ethyl-2-(4-methoxybenzylsulfanyl)-pyrimidin-4(3H)-one (**5v**).* Yield 46%; recrystallized from MEK as a colorless block crystal; mp: 231.2–231.5 °C; FT-IR (KBr): *ν* 3407 (NH), 2968, 2934, 2867, 2933 (CH_3, CH_2), 1635 (C=O), 1572, 1548, 1511, 1455 (aryl) cm^{-1}; ^1H NMR (DMSO-d_6): *δ* (ppm) 1.10 (t, 3H, $J = 7.6$ Hz, CH_2CH_3), 2.55 (q, 2H, $J = 7.6$ Hz, CH_2CH_3), 3.70 (s, 3H, OCH_3), 3.94 (s, 2H, $ArCH_2$), 4.25 (s, 2H, SCH_2), 6.70 (d, 2H, $J = 8.4$, $Ar'H_{3,5}$), 6.79 (d, 2H, $J = 8.4$, $Ar'H_{2,6}$), 7.25 (t, 1H, $J = 8.0$ Hz ArH_4), 7.47 (d, 2H, $J = 8.0$ Hz, $ArH_{3,5}$), 12.53 (br s, 1H, CONH); ^{13}C NMR (DMSO-d_6): *δ* (ppm) 12.67 (CH_2CH_3), 18.07 (CH_2CH_3), 32.27 (CH_2), 34.96 (CH_2), 55.17 (OCH_3), 113.80 (2C), 121.04 (C-5), 128.23 (2C), 129.23, 129.79 (2C), 129.96, 134.75, 135.92 (2C), 157.20 (C-6), 157.53 (Ar′-C_4), 158.46 (C-2), 162.82 (C-4); MS (EI): *m/z* 434 (M^+); Anal. Calcd for $C_{21}H_{20}Cl_2N_2O_2S$ (434.06): C, 57.93; H, 4.63; N, 6.43; S, 7.37. Found: C, 57.95; H, 4.60; N, 6.43; S, 7.36%.

*4.1.3.23. 6-(2,6-Dichlorobenzyl)-5-ethyl-2-(4-nitrobenzylsulfanyl)-pyrimidin-4(3H)-one (**5w**).* Yield 44%; column chromatography (eluent CH_2Cl_2/AcOEt 100/10), white solid; mp: 212.0–212.6 °C; FT-IR (KBr): *ν* 3423 (NH), 2977, 2938, 2867 (CH_3, CH_2), 1655 (C=O), 1561, 1519, 1434 (aryl) cm^{-1}; ^1H NMR (DMSO-d_6): *δ* (ppm) 1.10 (t, 3H, $J = 7.6$ Hz, CH_2CH_3), 2.56 (q, 2H, $J = 7.6$ Hz, CH_2CH_3), 4.14 (s, 2H, $ArCH_2$), 4.25(s, 2H, SCH_2), 7.10 (d, 2H, $J = 8.4$ Hz, $Ar'H_{2,6}$), 7.22 (t, 1H, $J = 8.0$ Hz, ArH_4), 7.42 (d, 2H, $J = 8.0$ Hz, $ArH_{3,5}$), 8.0 (d, 2H, $J = 8.4$, $Ar'H_{3,5}$), 12.68 (br s, 1H, CONH); MS (EI) *m/z* 449 (M^+); Anal. Calcd for $C_{20}H_{17}Cl_2N_3O_3S$ (449.04): C, 53.34; H, 3.80; N, 9.33; S, 7.12. Found: C, 53.34; H, 3.81; N, 9.35; S, 7.11%.

*4.1.3.24. 5-Ethyl-2-(4-methoxybenzylsulfanyl)-6-(1-naphthylmethyl)-pyrimidin-4(3H)-one (**5x**).* Yield 28%; column chromatography (eluent CH_2Cl_2/AcOEt, 100/5–100/25), white solid; mp: 166.6–168.1 °C; FT-IR (KBr): *ν* 3397 (NH), 2973, 2935, 2834 (CH_3, CH_2), 1646 (C=O), 1611, 1569, 1547, 1511 (aryl) cm^{-1}; ^1H NMR ($CDCl_3$): *δ* (ppm) 1.02 (t, 3H, $J = 7.6$ Hz, CH_2CH_3), 2.62 (q, 2H, $J = 7.6$ Hz, CH_2CH_3), 3.71 (s, 3H, OCH_3), 3.99 (s, 2H, $ArCH_2$), 4.38 (s, 2H, SCH_2), 6.57 (d, 2H, $J = 8.8$ Hz, $Ar'H_{3,5}$), 6.82 (d, 2H, $J = 8.8$ Hz $Ar'H_{2,6}$), 7.22–8.07 (m, 7H, ArH), 12.74 (br s, 1H, CONH); ^{13}C NMR ($CDCl_3$): *δ* (ppm) 13.03 (CH_2CH_3), 18.79 (CH_2CH_3), 33.74 (CH_2), 37.46 (CH_2), 55.13 (OCH_3), 113.68 (2C), 122.67 (C-5), 124.07, 125.43, 125.54, 125.95, 126.92, 127.20, 128.65, 128.99, 130.02 (2C), 132.42, 133.80, 134.39, 156.38 (C-6), 158.68 (Ar′-C_4), 161.42 (C-2), 165.05 (C-4); MS (EI): *m/z* 416 (M^+); Anal. Calcd for $C_{25}H_{24}N_2O_2S$ (416.16): C, 72.09; H, 5.81; N, 6.73; S, 7.70. Found: C, 72.11; H, 5.78; N, 6.76; S, 7.71%.

*4.1.3.25. 2-(4-Methoxybenzylsulfanyl)-6-(1-naphthylmethyl)-5-(isopropyl)pyrimidin-4(3H)-one (**5y**).* Yield 14%; column chromatography (petroleum ether/acetone 100/20), yellow solid; mp: 151.2–151.6 °C; FT-IR (KBr): *ν* 3420 (NH), 2962, 2930, 2867, 2836 (CH_3, CH_2), 1647 (C=O), 1560, 1541, 1510, 1459 (aryl) cm^{-1}; ^1H NMR ($CDCl_3$): *δ* (ppm) 1.31 (d, 6H, $J = 7.2$ Hz, $CH(CH_3)_2$), 3.07 (sept, 1H, $J = 7.2$ Hz, $CH(CH_3)_2$), 3.73 (s, 3H, OCH_3), 4.05 (s, 2H, $ArCH_2$), 4.43 (s, 2H, SCH_2), 6.61 (d, 2H, $J = 8.4$ Hz, $Ar'H_{3,5}$), 6.92 (d, 2H, $J = 8.4$ Hz, $Ar'H_{2,6}$), 7.16–8.10 (m, 7H, ArH), 12.59 (br s, 1H, CONH); ^{13}C NMR ($CDCl_3$): *δ* (ppm) 19.76 (2C, $2 \times CH_3$), 28.00 (CH), 33.76 (CH_2), 38.03 (CH_2), 55.18 (OCH_3), 113.74 (2C), 123.91 (C-5), 125.46, 125.59, 125.70, 125.98, 126.46, 127.13, 128.71, 129.10, 130.10 (2C), 132.29, 133.81, 134.54, 156.46 (C-6), 158.74 (Ar′-C_4), 160.81 (C-2), 164.37 (C-4); MS (EI): *m/z* 430 (M^+); Anal. Calcd for $C_{26}H_{26}N_2O_2S$ (430.17): C, 72.53; H, 6.09; N, 6.51; S, 7.45. Found: C, 72.51; H, 6.06; N, 6.55; S, 7.49%.

4.2. Anti-HIV assays

The activity of the compounds against HIV-1 (HTLV-III_B strain) and HIV-2 (ROD) was based on the inhibition of virus-induced cytopathic effect in MT-4 cells using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) method [13]. 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 dimethyl sulfoxide at 50 mM or higher. 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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