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Synthesis and Anti-HIV-1 Activity of Novel MKC-442 Analogues Containing Alkenyl Chains or Reactive Functionalities in the 6-Benzyl Group

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Summary. In an effort to obtain more insight into the anti-HIV efficacy of MKC-442 analogues (1-(alkoxymethyl)-6-benzyluracils), a new series of compounds was synthesized and evaluated for inhibition of HIV-1 replication. The modifications include a reactive center such as an aldehyde or an epoxide substituted at the benzyl group. It was believed that such reactive groups could improve the activity against HIV for the Y181C mutant by forming a covalent bond to the mercapto group in cysteine in the hydrophobic pocket. Unfortunately, only moderate activities were found in cell-based assays for such compounds against wild-type HIV and no activity against the Y181C mutant. However, higher activities were found for a corresponding oxime and the precursor molecules with butenyl and allyloxy substituents in the benzyl group. A few amino-*DABO* and *S-DABO* analogues were also synthesized, but they were found inactive against HIV.

Keywords. Bioorganic chemistry; Drug research; HIV-1; MKC-442; Non-nucleoside.

Introduction

A major challenge facing medicinal chemistry over the next few years will be the development of drugs with significantly improved resistance profiles for chronic use in anti-HIV combination therapy. An important component of such regimens

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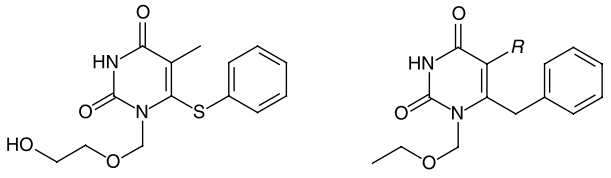


Fig. 1. Non-nucleoside reverse transcriptase inhibitors of HIV-1

will be non-nucleoside reverse transcriptase inhibitors (NNRTIs) of HIV-1. This is a class of structurally diverse aromatic compounds, such as nevirapine, delavirdine, efavirenz, MKC-442 (emivirine or coactinon), *HEPT* (1-[(2-hydroxyethoxy)methyl]-6-(phenylthio)thymine, **1**, Fig. 1), TIBO, and thiocarboxanilides [1]. Structural studies have revealed that NNRTIs inhibit HIV-1 reverse transcriptase (RT) by binding to an allosteric site, approximately 10 Å from the polymerase active site [2–4] causing a distortion of the catalytic aspartate triad [5].

MKC-442 (**2a**, Fig. 1) is an optimized structure of *HEPT* which was abandoned during phase III clinical trials [6]. Also 2,6-disubstituted and 2,5,6-trisustituted-4(3*H*)-pyrimidinones (amino-*DABO*s and *S-DABO*s) [7, 8] are antiviral agents inhibiting HIV-1 RT [9]. In an attempt to optimize the MKC-442 lead structure we have previously introduced a vinyl group at C-5 of the uracil ring (**2b**, Fig. 1) instead of the isopropyl group [10]. This compound has shown good activity against HIV-1 ($IC\text{-}50 = 0.07\,\mu M$) being almost as active as MKC-442 ($IC\text{-}50 = 0.02\,\mu M$).

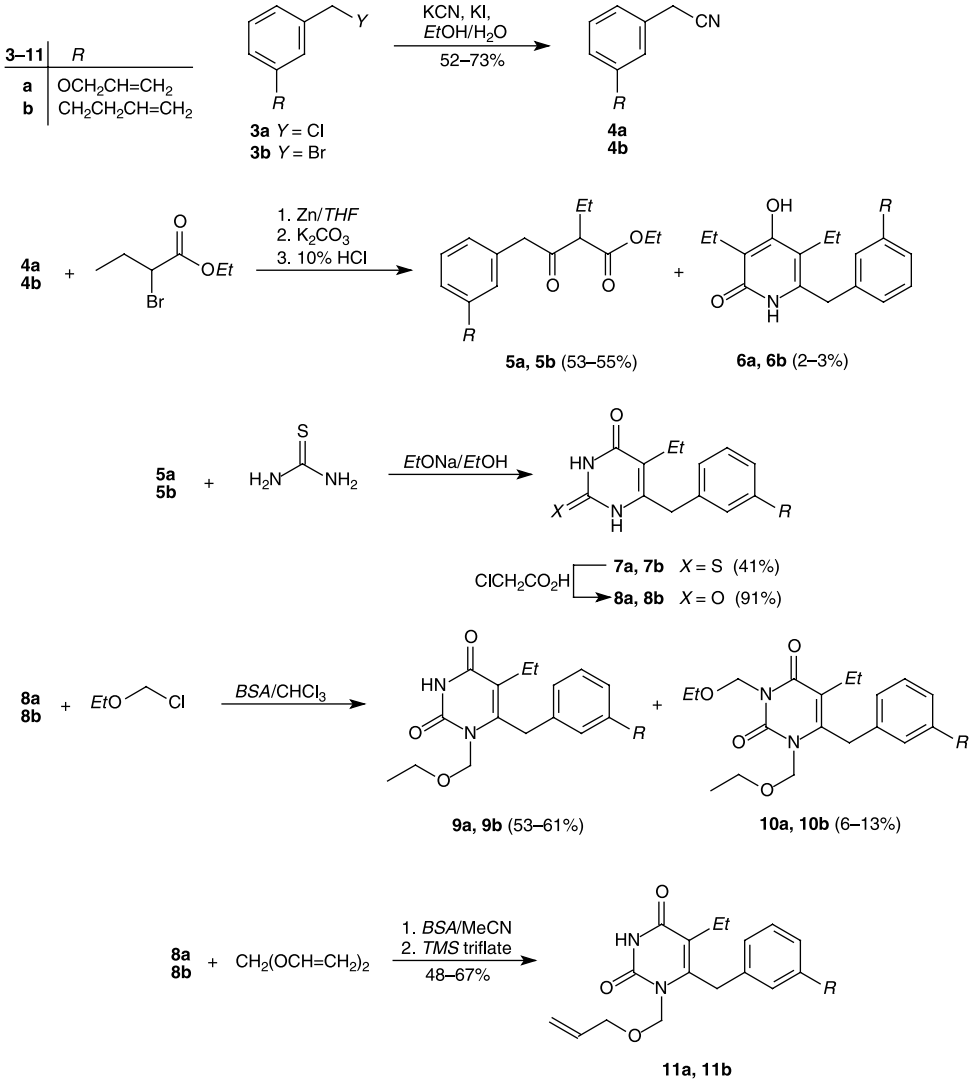
When NNRTIs are used as drugs against HIV the major problem is the fast development of resistance. A very common mutation when using MKC-442 is the conversion of tyrosine at position 181 in RT into cysteine (Y181C). We have previously reported the synthesis of an MKC-442 analogue with a carbaldehyde at position C-5 [10]. It was hoped that the drug would covalently bind to the hydrophobic pocket by formation of an a thiohemiacetal between the carbaldehyde and the mercapto group in cysteine of the mutated virus. This carbaldehyde turned out to be inactive toward the wild-type as well as the mutant. We have also attempted to synthesize an epoxide directly attached to C-5 and also further away from the uracil ring. However, the first compounds turned out to be too reactive to be isolated, its ring being opened by the *m*-chlorobenzoic acid formed during epoxidation of the corresponding alkene, the second one has been isolated [10, 11]. In this work we have introduced the same reactive groups in the aromatic ring instead of the uracil ring hoping for a more appropriate position of the reactive groups for reaction with the mercapto group in the Y181 mutation of RT. A flexible linker to the reactive groups from the aromatic ring may further improve the chance of a reaction with the mercapto group.

Results and Discussion

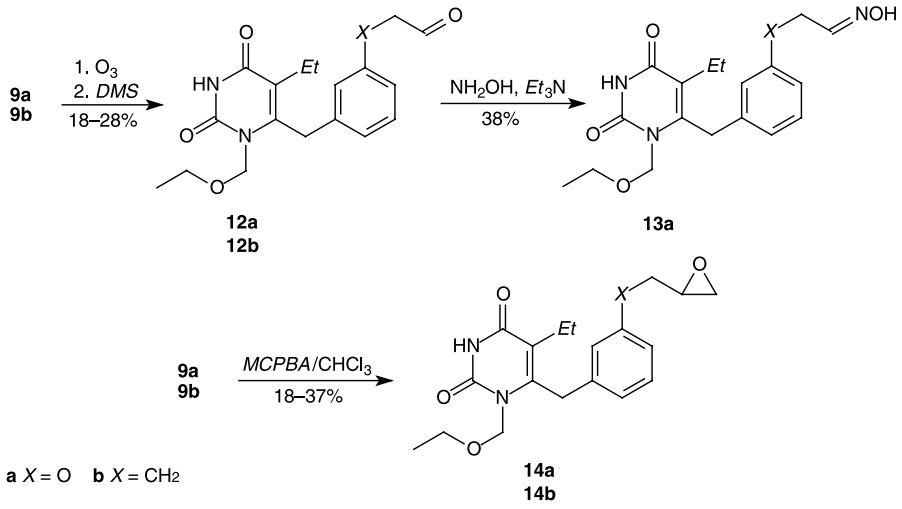
Chemistry

m-(Allyloxy)benzyl chloride (**3a**) was synthesized from reaction of *m*-hydroxybenzyl alcohol with allyl bromide in a successive alkylation reaction and chloronation

with thionyl chloride [12]. The corresponding *m*-butenylbenzyl bromide (**3b**) was synthesized by reaction of *m-bis*(bromomethyl)benzene with allylmagnesium bromide in 43% yield. Refluxing **3a**, **3b** with KCN in aqueous ethanol in the presence of KI furnished the nitriles **4a**, **4b** in 52–73% yield. Reaction of **4a**, **4b** with the zinc organometallic reagent prepared from ethyl 2-bromobutyrate in anhydrous *THF* afforded the β-ketoesters **5a**, **5b** in 53–55% yield and the pyridinones **6a**, **6b** as by-products. Compounds **5a**, **5b** were used as starting materials for the synthesis of uracil rings in a ring closure reaction with thiourea and NaO*Et*. The formed thiouracils **7a**, **7b** were desulfurized with chloroacetic acid [13, 14] to give the uracil derivatives **8a**, **8b** in overall yield of 37% in both cases for the two steps from **5a**, **5b**. Compounds **8a**, **8b** were silylated using bis(trimethylsilyl)acetamide (*BSA*) and subsequently N-1 alkylated using chloromethyl ethyl ether [13] to give



Scheme 1

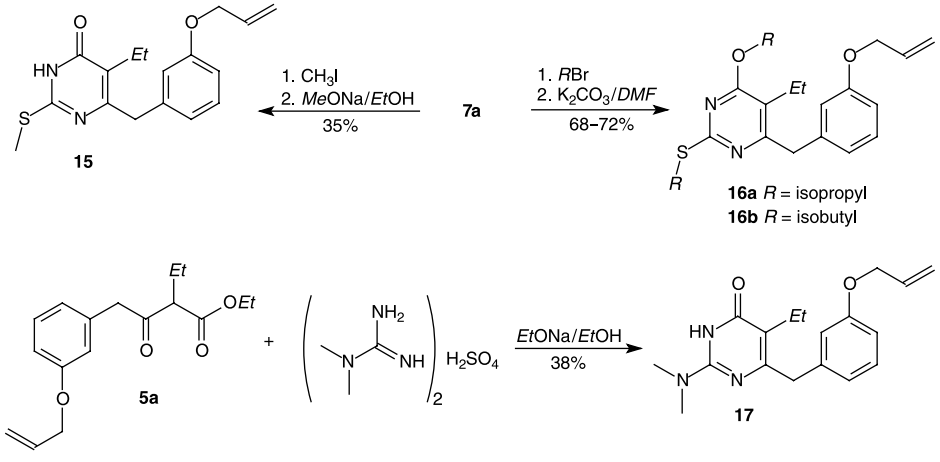


Scheme 2

the non-nucleoside analogues **9a**, **9b** in 53–61% yield and the N-1,N-3 bisalkylated by-products **10a**, **10b** in 6–13% yield. The uracils **8a**, **8b** were also silylated and reacted with bis(allyloxy)methane in the presence of trimethylsilyl trifluoromethanesulfonate (*TMS* triflate) as a *Lewis* acid catalyst [15] to give the non-nucleoside analogues **11a**, **11b** in 48–67% yield (Scheme 1).

The uracil derivatives **9a**, **9b** were reacted with ozone to cleave the double bond and the aldehydes **12a**, **12b** were isolated in 18–28% yield and compound **12a** was converted to the corresponding oxim **13a** in 38% yield. In order to epoxidize the double bond, the compounds **9a**, **9b** were reacted with *m*-chloroperbenzoic acid (*MCPBA*) in dry $CHCl_3$ to give the epoxides **14a**, **14b** in 18–37% yield (Scheme 2).

Alkylation of **7a** with isopropyl bromide or isobutyl bromide in the presence of K_2CO_3 in dry *DMF* gave *O*- and *S*-bisalkylated products **16a**, **16b**. The mono *S*-alkylated derivatives were not observed. When sodium methoxide and excess



Scheme 3

methyl iodide were used, the *S*-alkylated product **15** was obtained without any bisalkylated products. For the synthesis of amino-*DABO* derivatives the β-ketoester **5a** was reacted with dimethylguanidine sulphate in *Et*ONa to afford compound **17** in 38% yield (Scheme 3).

Antiviral Activity

The anti-HIV activities and cytotoxicities of the synthesized MKC-442 analogues are summarized in Table 1. In general, the introduction of an allyloxy or a butenyl group in the benzylic part at the 3-position did not improve the activity against HIV-1 compared to MKC-442. In fact that compounds **9a**, **9b** and **11a**, **11b** showed a 10-fold lower activity against the wild-type HIV-1 than MKC-442. Therefore it was surprising to find better activities for these compounds against the mutant Y181C than the one found for MKC-442. Even against the double mutant, activity was observed for the derivatives **9b** and **11b**. The compounds containing an oxim **13a**, a reactive aldehyde group **12a**, **12b** or an reactive epoxides **14a**, **14b** in the

Table 1. Cytotoxicity and anti-HIV-1 activity of compounds **6–17**[a]

	Wild-type		Mutants/μM		
	CC-50[b]/μM	*IC*-50[c]/μM	EFV^R	Y181C	K103N + Y181C
6a	77	>20	>20	>20	>20
6b	95 ± 5	>95	>95	>95	>95
7a	86	>20	>20	>20	>20
7b	80 ± 18	>80	>80	>80	>80
8a	74	>20	>20	>20	>20
8b	66 ± 4	>66	>66	>66	>66
9a	65 ± 6	0.5 ± 0.05	>65	7 ± 3	>65
9b	55 ± 5	0.3 ± 0.1	>55	2 ± 0.2	10 ± 1
10a	41	>20	>20	>20	>20
10b	45 ± 5	>45	>45	>45	>45
11a	44 ± 2	0.4 ± 0.005	20	4 ± 0.8	>44
11b	42 ± 2	0.2 ± 0.05	30 ± 8	1 ± 0.2	5 ± 2
12a	45 ± 1	8 ± 1	>45	15 ± 5	>45
12b	>100	1.9 ± 0.05	>100	51 ± 8	>100
13a	>100	0.3 ± 0.1	>100	35 ± 5	54
14a	80 ± 20	14 ± 3	>80	>80	>80
14b	>100	9 ± 1	>100	80 ± 20	80 ± 20
15	80 ± 20	>80	>80	>80	>80
16a	>100	>100	>100	>100	>100
16b	>100	>100	>100	>100	>100
17	>100	>20	>20	>20	>20
MKC-442	>100	0.03	100	20	>100

[a] Data represent mean values of at least two separate experiments; [b] compound dose required to reduce the viability of mock-infected cells by 50%, as determined by the *MTT* method; [c] compound dose required to achieve 50% protection of MT-4 cells from HIV-1-induced cytopathogenicity, as determined by the *MTT* method

side chain of the benzylic group showed rather moderate activities against the wild-type virus when compared to MKC-442. Also, they do not show activity against the mutants Y181C and K103N + Y181C. The latter indicates that the covalent binding of the drug to cysteine in the RT hydrophobic pocket has not taken place or that this principle is not a sufficient prerequisite for activity against the HIV mutants. The examples of *S-DABO* **15** and amino-*DABO* **17** with an 3-allyloxy substitutent in the benzylic part were also devoid of activity against HIV-1.

Experimental

NMR spectra were recorded on a Varian Gemini 2000 spectrometer at 300 MHz for ^{1}H and 75 MHz for ^{13}C NMR, internal standards used in ^{1}H NMR spectra were *TMS*. Accurate-ion mass determinations were performed using the 4.7 T Ultima *Fourier* transform mass spectrometer (Ion Spec, Irvine, CA). The $(M+H)^+$ and $(M+Na)^+$ ions were peak matched using ions derived from the 2,5-dihydroxybenzoic acid matrix. Elemental analyses were performed at H.C. Ørsted Institute, University of Copenhagen; the found values agreed favourably with the calculated ones. Thin-layer chromatography (TLC) analyses were carried out with Silica Gel 60 F_{254} TLC plates purchased from E. Merck and were visualized in UV light (254 nm). The silica gel (0.040–0.063 mm) used for column chromatography was purchased from E. Merck. Solvents used for column chromatography were distilled prior to use, while reagents were used as purchased.

m-Butenylbenzyl bromide (**3b**)
To *m*-bis(bromomethyl)benzene (1.35 g, 5.14 mmol) in dry ether (30 cm^3), was added dropwise allylmagnesium chloride (5.14 mmol) in 20 cm^3 dry ether at 0°C. After complete addition the reaction mixture was refluxed for 20 h. The solvent was evaporated and the residue was chromatographed using petroleum ether (60–80°C)/ethyl acetate (1/1, *v*/*v*) to afford compound **3b**. Yield 43%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 2.34$–2.39 (m, 2H, CH_2CH), 2.66–2.71 (m, 2H, CH_2Ph), 4.45 (s, 2H, CH_2Br), 5.05–5.06 (m, 2H, CH_2=CH), 5.82–5.88 (m, 1H, CH=CH_2), 7.09–7.24 (m, 4H, H_{arom}) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 33.68$ (*C*H_2CH), 35.10 (CH_2Ph), 35.24 (CH_2Br), 115.07 (*C*H_2=CH), 137.69 (*C*H=CH_2), 125.84, 126.49, 128.52, 128.67, 129.04, 142.42 (C_{arom}) ppm; MS (EI): $m/z = 224$ (M^+).

*General Procedure for Preparation of Compounds **4a** and **4b***
To compound **3a** or **3b** (150 mmol) in 100 cm^3 ethanol, were added KCN (22 g, 340 mmol) and KI (5 g, 30 mmol) in 100 cm^3 H_2O. The reaction mixture was refluxed for 4 h, cooled to room temperature, filtered, and the ethanol was evaporated. The residue was dissolved in H_2O and extracted with diethyl ether. The organic layer was dried over $MgSO_4$ and evaporated and the residue was chromatographed (petroleum ether (60–80°C)/ethyl acetate = 8/2, *v*/*v*) to afford the compounds **4a** and **4b**.

1-Allyloxy-3-cyanomethylbenzene (**4a**)
Yield 73%; oil; IR: $\bar{\nu} = 2251$ (CN) cm^{-1}; ^{1}H NMR (300 MHz, *DMSO*-d_6): $\delta = 3.98$ (s, 2H, CH_2CN), 4.56 (d, $J = 2.1$ Hz, 1H, CH_2O), 5.36–5.42 (m, 2H, CH_2=CH), 6.01–6.05 (m, 1H, CH=CH_2), 6.93–7.33 (m, 4H, H_{arom}) ppm; ^{13}C NMR (75 MHz, *DMSO*-d_6): $\delta = 22.28$ (*C*H_2CN), 68.09 (CH_2O), 117.9 (*C*H_2=CH), 119.06 (CN), 132.79 (*C*H=CH_2), 114.19, 114.37, 120.21, 130.07, 131.20, 159.01 (C_{arom}) ppm; MS (EI): $m/z = 173$ (M^+).

1-(But-3-enyl)-3-cyanomethylbenzene (**4b**)
Yield 52%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 2.35$–2.37 (m, 2H, CH_2CH), 2.68–2.73 (t, $J = 7.7$ Hz, 2H, CH_2Ph), 3.70 (s, 2H, CH_2CN), 4.99–5.00 (m, 2H, CH_2=CH), 5.78–5.84 (m, 1H, CH=CH_2), 7.12–7.30 (m, 4H, H_{arom}) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 23.43$ (*C*H_2CN), 35.22

(CH_2CH), 36.04 (CH_2Ph), 115.14 (CH_2=CH), 117.89 (CN), 137.52 (CH=CH_2), 125.32, 127.90, 128.05, 128.95, 129.75, 142.87 (C_{arom}) ppm; MS (EI): $m/z = 171$ (M^+).

*General Procedure for Preparation of Compounds **5a**, **5b** and **6a**, **6b***

Zn dust (25–30 g) was activated by stirring with 4 *M* HCl (100 cm^3) for 5 min. The zinc dust was filtered off and washed sequentially with H_2O, ethyl alcohol, and ether, dried by evaporation under reduced pressure at 80°C for 5 h, and kept *in vacuo* overnight. The active Zn was suspended in dry *THF* (150 cm^3) and heated to reflux. A few drops of ethyl 2-bromobutyrate were added and the mixture was refluxed until a green colour appeared. Compound **4a** or **4b** (66 mmol) was added in one portion and ethyl 2-bromobutyrate (25.3 g, 130 mmol) in 10 cm^3 *THF* was added dropwise. After complete addition the mixture was refluxed for 5 h. After cooling and dilution with *THF* (300 cm^3), the mixture was quenched by addition of sat aqueous K_2CO_3 (100 cm^3). The mixture was stirred for 1 h and then the *THF* layer was decanted and the residue was washed with *THF* (3×100 cm^3). The combined *THF* fractions were stirred with 10% aqueous HCl (90 cm^3) for 45 min, then the solution was concentrated under reduced pressure and CH_2Cl_2 (300 cm^3) was added. The organic phase was washed with sat aqueous $NaHCO_3$ (2×100 cm^3), dried over $MgSO_4$, and evaporated under reduced pressure. The residue was chromatographed on silica gel (petroleum ether (60–80°C)/ethyl acetate = 80/20, *v*/*v*) to give **5a**, **5b** and **6a**, **6b**.

Ethyl 2-ethyl-3-oxo-4-[3-(allyloxy)phenyl]butyrate (**5a**)

Yield 55%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 0.84$–0.92 (m, 3H, C$H_3$$CH_2$), 1.22–1.24 (m, 3H, C$H_3$$CH_2O$), 1.84–1.88 (m, 2H, C$H_2$$CH_3$), 3.44–3.47 (m, 1H, COCHCO), 3.77 (s, 2H, PhCH_2), 4.14–4.17 (m, 2H, OC$H_2$$CH_3$), 4.50–4.52 (m, 2H, OC$H_2$CH), 5.29–5.37 (m, 2H, C$H_2$=CH), 6.01–6.08 (m, 1H, CH=CH_2), 6.76–7.26 (m, 4H, H_{arom}) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 11.74$, 14.03 (2s, $2CH_3$), 21.46 (CH_2CH_3), 49.03 ($CH_2 \cdot Ph$), 59.36 (CO*C*HCO), 61.24 (OCH_2CH_3), 68.67 (PhOCH_2), 117.57 (CH_2=CH), 133.10 (*C*H=CH_2), 113.41, 116.03, 122.07, 129.57, 134.64, 158.74 (C_{arom}), 169.52 (*C*OCH_2Ph), 202.43 (*C*OOCH_2) ppm; MS (EI): $m/z = 290$ (M^+).

Ethyl 2-ethyl-3-oxo-4-[3-(but-3-enyl)phenyl]butyrate (**5b**)

Yield 53%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 0.81$–0.85 (m, 3H, C$H_3$$CH_2$), 1.22–1.26 (m, 3H, C$H_3$$CH_2O$), 1.83–1.88 (m, 2H, C$H_2$$CH_3$), 2.34–2.39 (m, 2H, C$H_2$CH), 2.66–2.71 (m, 2H, C$H_2$$CH_2$), 3.46 (t, $J = 7.4$ Hz, 1H, COCHCO), 3.78 (s, 2H, Ph · CH_2CO), 4.11–4.16 (m, 2H, OC$H_2$$CH_3$), 4.98–5.00 (m, 2H, C$H_2$=CH), 5.79–5.88 (m, 1H, CH=CH_2), 7.02–7.26 (m, 4H, H_{arom}) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 11.74$, 14.04 (2s, $2CH_3$), 21.47 (CH_2CH_3), 35.17 (Ph$CH_2$$CH_2$), 35.37 ($CH_2$CH), 49.06 (CO$CH_2$Ph), 59.35 (CO*C*HCO), 61.20 (O$CH_2$$CH_3$), 114.93 (*C*$H_2$=CH), 133.15 (*C*H=$CH_2$), 127.07, 127.24, 128.55, 129.68, 137.83, 142.26 (C_{arom}), 169.54 (*C*OCH_2Ph), 202.64 (*C*OOCH_2) ppm; MS (EI): $m/z = 288$ (M^+).

6-[3-(Allyloxy)benzyl]-4-hydroxy-3,5-diethylpyridin-2(1H)-one (**6a**, $C_{19}H_{23}NO_3$)

Yield 2%; mp 188–190°C; ^{1}H NMR (300 MHz, *DMSO*-d_6): $\delta = 0.84$–0.97 (m, 6H, $2CH_3$), 2.36–2.51 (m, 4H, $2CH_2$), 3.78 (s, 2H, PhCH_2), 4.50 (d, $J = 5.18$ Hz, 2H, OCH_2), 5.21–5.47 (m, 2H, CH_2=CH), 5.94–6.07 (m, 1H, CH=CH_2), 6.76–7.26 (m, 4H, H_{arom}) 9.11–9.29 (br.s, 1H, OH), 11.04–11.12 (br.s, 1H, NH) ppm; ^{13}C NMR (75 MHz, *DMSO*-d_6): $\delta = 13.10$, 14.30 (2s, $2CH_3$), 16.04, 17.97 ($2CH_2$), 35.02 (CH_2Ph), 68.04 (PhOCH_2), 117.40 (CH_2=CH), 133.61 (*C*H=CH_2), 111.03 (C-3), 111.33 (C-5), 112.15, 114.88, 120.46, 129.43, 139.84, 139.98 (C_{arom}), 158.18 (C-6), 161.64 (C-4), 163.22 (C=O) ppm.

6-[3-(But-3-enyl)benzyl]-4-hydroxy-3,5-diethylpyridin-2(1H)-one (**6b**, $C_{20}H_{25}NO_2$)

Yield 3%; mp 105–107°C; ^{1}H NMR (300 MHz, *DMSO*-d_6): $\delta = 0.78$–0.93 (m, 6H, $2CH_3$), 2.22–2.58 (m, 8H, $4CH_2$), 3.75 (s, 2H, PhCH_2), 4.93–4.99 (m, 2H, CH_2=CH), 5.74–5.82 (m, 1H, CH=CH_2), 6.98–7.17 (m, 4H, H_{arom}), 9.15–9.18 (br.s, 1H, OH), 11.18–11.20 (br.s, 1H, NH)

ppm; ^{13}C NMR (75 MHz, *DMSO*-d_6): $\delta = 13.06$, 14.20 (2s, $2CH_3$), 16.02, 17.95 ($2CH_2CH_3$), 34.39 ($PhCH_2CH_2$), 34.84 (CH_2Ph), 35.10 (CH_2CH), 115.10 (CH_2=CH), 137.82 (CH=CH_2), 110.93 (C-3), 111.25 (C-5), 125.51, 126.27, 128.18, 128,27, 138.32, 140.06, (C_{arom}), 141.49 (C-4), 161.16 (C-6), 163.26 (C=O) ppm.

General Procedure for Preparation of 5,6-Substituted 2-Thioxo-2,3-dihydro-1H-pyrimidin-4-one ***7a*** *and* ***7b***

Na (25.1 g, 1.1 mol) was dissolved in 500 cm^3 absolute ethanol. Thiourea (58.23 g, 0.77 mol) was added and the mixture was heated to reflux. Compound **5a** or **5b** (0.015 mol) was added dropwise, and the mixture was refluxed for 5 h. Ethanol was evaporated *in vacuo* and the residue was dissolved in 400 cm^3 H_2O. The thiouracil **7a** or **7b** was precipitated by neutralization with conc HCl. The precipitate was filtered off, washed with H_2O, and chromatographed on silica gel (petroleum ether (60–80°C)/ethyl acetate = 1/1, *v*/*v*) to give **7a** and **7b**.

6-[3-(Allyloxy)benzyl]-5-ethyl-2-thioxo-2,3-dihydro-1H-pyrimidin-4-one (**7a**)

Yield 41%; mp 69–71°C; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 0.81$–0.86 (m, 3H, CH_3CH_2), 2.26–2.29 (m, 2H, CH_2CH_3), 3.81 (s, 2H, CH_2Ph), 4.53 (d, $J = 4.9$ Hz, 2H, OCH_2), 5.35–5.40 (m, 2H, CH_2=CH), 6.00–6.18 (m, 1H, CH=CH_2), 6.78–7.27 (m, 4H, H_{arom}), 12.21, 12.41 (2s, 2H, 2NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 12.82$ (CH_3), 17.67 (CH_2CH_3), 34.42 (CH_2Ph), 68.06 ($PhOCH_2$), 112.62 (C-5), 117.40 (CH_2=CH), 133.65 (CH=CH_2), 114.89, 117.05, 120.35, 129.64, 138.16, 149.02 (C_{arom}), 158.23 (C-6), 161.38 (CO), 174.11 (C=S) ppm; MS (MALDI): $m/z = 325$ ($M + Na^+$).

6-[3-(But-3-enyl)benzyl]-5-ethyl-2-thioxo-2,3-dihydro-1H-pyrimidin-4-one (**7b**, $C_{17}H_{20}N_2OS$)

Yield 41%; mp 179–181°C; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 1.00$–1.05 (t, $J = 7.4$ Hz, 3H, CH_3CH_2), 2.32–2.47 (m, 4H, CH_2CH_3, CH_2CH_2), 2.65–2.70 (q, $J = 7.6$ Hz, 2H, CH_2CH), 3.84 (s, 2H, CH_2Ph), 4.98–5.01 (m, 2H, CH_2=CH), 5.86–5.88 (m, 1H, CH=CH_2), 6.98–7.26 (m, 4H, H_{arom}), 10.47, 10.51 (2s, 2H, 2NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.12$ (CH_3), 18.23 (CH_2CH_3), 35.11 (CH_2CH), 35.25 (CH_2CH_2Ph), 35.58 (CH_2Ph), 115.18 (C-5), 118.40 (CH_2=CH), 137.62 (CH=CH_2), 125.92, 127.88, 128.59, 129.14, 134.09, 142.97 (C_{arom}), 149.15 (C-6), 161.88 (CO), 173.83 (C=S) ppm; HRMS (MALDI, peak matching): $m/z = 323.1193$ ($M + Na^+$) calcd 323.1189.

General Procedure for Preparation of 5,6-Substituted 1H-Pyrimidine-2,4-dione ***8a*** *and* ***8b***

Compound **7a** or **7b** (0.01 mol) was suspended in 300 cm^3 10% aqueous chloroacetic acid. The suspension was refluxed overnight and the precipitate was filtered off after cooling. The precipitate was washed with H_2O and dried *in vacuo* to give compounds **8a** and **8b**.

6-[3-(Allyloxy)benzyl]-5-ethyl-1H-pyrimidin-2,4-dione (**8a**, $C_{16}H_{18}N_2O_3$)

Yield 91%; mp 155–157°C; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 0.81$–0.86 (m, 3H, CH_3CH_2), 2.24–2.26 (m, 2H, CH_2CH_3), 3.71 (s, 2H, CH_2Ph), 4.53 (d, $J = 1.4$ Hz, 2H, OCH_2), 5.22–5.26 (m, 2H, CH_2=CH), 6.00–6.04 (m, 1H, CH=CH_2), 6.81–7.26 (m, 4H, H_{arom}), 10.72, 11.01 (2s, 2H, 2NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.46$ (CH_3), 17.57 (CH_2CH_3), 34.92 (CH_2Ph), 68.07 ($PhOCH_2$), 111.27 (C-5), 117.39 (CH_2=CH), 133.60 (CH=CH_2), 112.63, 114.84, 120.41, 129.61, 138.33, 148.52 (C_{arom}), 150.88 (C-6), 158.24, 164.45 (2CO) ppm; HRMS (MALDI, peak matching): $m/z = 309.1213$ ($M + Na^+$) calcd 309.1384.

6-[3-(But-3-enyl)benzyl]-5-ethyl-1H-pyrimidin-2,4-dione (**8b**, $C_{17}H_{20}N_2O_2$)

Yield 91%; mp 182–184°C; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 1.01$–1.05 (m, 3H, CH_3CH_2), 2.30–2.35 (m, 2H, CH_2CH_3), 2.45–2.68 (m, 4H, $PhCH_2CH_2$, CH_2CH), 3.76 (s, 2H, CH_2Ph), 4.92–5.02 (m, 2H, CH_2=CH), 5.75–5.84 (m, 1H, CH=CH_2), 7.04–7.25 (m, 4H, H_{arom}), 9.88 (s, 2H, 2NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.60$ (CH_3), 18.15 (CH_2CH_3), 35.10 (CH_2CH), 35.25 (CH_2CH_2Ph), 36.02 (CH_2Ph), 113.28 (C5), 115.04 (CH_2=CH), 137.69 (CH=CH_2), 126.11, 127.58, 128.75, 128.91,

134.89, 142.71 (C_{arom}), 148.65 (C-6), 151.98, 164.96 (2CO) ppm; HRMS (MALDI, peak matching): $m/z = 307.1418$ $(M + Na)^+$ calcd 307.1417.

*General Procedure for Synthesis of **9a**, **9b** and **10a**, **10b***

N,O-Bis(trimethylsilyl)acetamide (*BSA,* 2.5 mmol, 6.2 cm^3) was dissolved in 30 cm^3 dry $CHCl_3$ under nitrogen. Compound **8a** or **8b** (10 mmol) was added and after 10 min the mixture become a clear solution. Chloromethyl ethyl ether (15 mmol, 1.5 cm^3) was added and the reaction mixture was stirred overnight at room temperature under nitrogen, quenched with 25 cm^3 ice cold sat aqueous. $NaHCO_3$, and extracted with CH_2Cl_2 (2×50 cm^3). The organic layer was dried over $MgSO_4$ and evaporated under reduced pressure and the residue was purified by column chromatography using ethyl acetate/petroleum ether (60–80°C) (1/1, *v*/*v*) to give compounds **9a**, **9b** and **10a**, **10b**.

6-[3-(Allyloxy)benzyl]-1-ethoxymethyl-5-ethyl-1H-pyrimidin-2,4-dione (**9a**, $C_{19}H_{24}N_2O_4$)
Yield 53%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 1.05–1.22$ (m, 6H, 2CH_3CH_2), 2.45–2.52 (m, 2H, CH_2CH_3), 3.60–3.72 (m, 2H, OCH_2CH_3), 4.11 (s, 2H, CH_2Ph), 4.49 (d, $J = 1.40$ Hz, 2H, OCH_2CH), 5.30–5.42 (m, 2H, CH=CH_2), 5.46 (s, 2H, NCH_2O), 5.98–6.04 (m, 1H, CH=CH_2), 6.70–7.26 (m, 4H, H_{arom}), 8.75 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.72$, 14.99 (2CH_3), 19.09 (*C*H_2CH_3), 32.28 (*C*H_2Ph), 64.92 (O*C*H_2CH_3), 68.73 (PhO*C*H_2), 72.64 (NCH_2O), 112.94 (C-5), 117.86 (*C*H_2=CH), 132.90 (*C*H=CH_2), 114.09, 116.87, 119.75, 130.14, 136.90, 148.90 (C_{arom}), 151.94 (C-6), 159.18, 163.38 (2CO) ppm; MS (EI): $m/z = 344$ (M^+).

6-[3-(But-3-enyl)benzyl]-1-ethoxymethyl-5-ethyl-1H-pyrimidin-2,4-dione (**9b**, $C_{20}H_{26}N_2O_3$)
Yield 61%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 1.04–1.28$ (m, 6H, 2CH_3CH_2), 2.23–2.35 (m, 2H, CH_2CH_3), 2.47–2.71 (m, 4H, PhCH_2CH_2, CH_2CH), 3.60–3.65 (m, 2H, OCH_2CH_3), 4.13 (s, 2H, CH_2Ph), 4.98–5.00 (m, 2H, CH_2=CH), 5.12 (s, 2H, NCH_2O), 5.77–5.82 (m, 1H, CH=CH_2), 6.95–7.27 (m, 4H, H_{arom}), 9.97 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.70$, 14.98 (2s, 2CH_3), 19.08 (*C*H_2CH_3), 33.31 (*C*H_2CH), 35.10 (*C*H_2CH_2Ph), 35.31 (*C*H_2Ph), 64.90 (OCH_2), 72.62 (NCH_2O), 115.10 (C-5), 116.81 (*C*H_2=CH), 124.74, 127.33, 129.05, 135.11, 142.90 (C_{arom}), 137.60 (*C*H=CH_2), 149.17 (C-6), 152.06, 163.55 (2CO) ppm; HRMS (MALDI, peak matching): $m/z = 365.1830$ $(M + Na^+)$ calcd 365.1836.

6-[3-(Allyloxy)benzyl]-1,3-bis(ethoxymethyl)-5-ethyl-1H-pyrimidin-2,4-dione (**10a**)
Yield 6%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 1.05–1.25$ (m, 9H, 3CH_3), 2.45–2.50 (m, 2H, CH_2CH_3), 3.57–3.74 (m, 4H, 2OCH_2CH_3), 4.16 (s, 2H, CH_2Ph), 4.50 (d, $J = 1.29$ Hz, 2H, OCH_2CH=CH_2), 5.13 (s, 2H, N^3CH_2O), 5.26–5.30 (m, 2H, CH=CH_2), 5.47 (s, 2H, N^1CH_2O), 5.98–6.07 (m, 1H, CH=CH_2), 6.70–7.27 (m, 4H, H_{arom}) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.67$, 15.00, 15.15 (3CH_3), 19.69 (*C*H_2CH_3), 33.36 (*C*H_2Ph), 65.02, 65.86 (2O*C*H_2CH_3), 68.75 (PhO*C*H_2), 71.09 (N^3CH_2O), 73.45 (N^1CH_2O), 112.84 (C-5), 117.89 (*C*H_2=CH), 132.89 (*C*H=CH_2), 114.21, 116.13, 119.79, 130.14, 136.79, 147.63 (C_{arom}), 152.51 (C-6), 159.17, 162.66 (2CO) ppm; MS (EI): $m/z = 402$ (M^+).

6-[3-(But-3-enyl)benzyl]-1,3-bis(ethoxymethyl)-5-ethyl-1H-pyrimidin-2,4-dione
(**10b**, $C_{23}H_{32}N_2O_4$)
Yield 13%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 1.04–1.26$ (m, 9H, 3CH_3), 2.32–2.35 (m, 2H, CH_2CH_3), 2.48–2.70 (m, 4H, PhCH_2CH_2, CH_2CH), 3.57–3.74 (m, 4H, 2OCH_2CH_3), 4.12 (s, 2H, CH_2Ph), 4.97–4.98 (m, 2H, CH_2=CH), 5.13 (s, 2H, N^1CH_2O), 5.47 (s, 2H, N^3CH_2O) 5.76–5.82 (m, 1H, CH=CH_2), 6.92–7.27 (m, 4H, H_{arom}) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.67$, 15.00, 15.15 (3CH_3), 19.69 (*C*H_2CH_3), 33.39 (*C*H_2CH_2Ph), 35.10 (*C*H_2Ph), 35.31 (*C*H_2CH), 64.98, 65.84 (2OCH_2), 71.07, 73.42 (2NCH_2O), 115.10 (C-5), 116.04 (*C*H_2=CH), 124.74, 127.34, 127.36, 129.05, 135.11, 142.89 (C_{arom}), 137.59 (*C*H=CH_2), 147.89 (C-6), 152.54, 162.67 (2CO) ppm; HRMS (MALDI, peak matching): $m/z = 423.2237$ $(M + Na^+)$ calcd 423.2254.

*General Procedure for Synthesis of **11a** and **11b***

Compound **8a** or **8b** (3 mmol) was dissolved in 30 cm^3 CH_3CN under nitrogen and *BSA* (10.5 mmol, 2.6 cm^3) was added. The reaction mixture became clear after 15 min and after cooling to −50°C *TMS* triflate (3 mmol, 0.54 cm^3) was added followed by dropwise addition of bis(allyloxy)methane. The reaction mixture was stirred at room temperature for 30 h, quenched with a cold solution of sat aqueous $NaHCO_3$ (5 cm^3). The solvent was evaporated under reduced pressure and the residue was extracted with diethyl ether (3 × 50 cm^3), dried over $MgSO_4$, evaporated under reduced pressure, and chromatographed using ethyl acetate/petroleum ether (60–80°C) (1/1, *v*/*v*) to give **11a** and **11b**.

6-[3-(Allyloxy)benzyl]-1-allyloxymethyl-5-ethyl-1H-pyrimidin-2,4-dione (**11a**, $C_{20}H_{24}N_2O_4$)

Yield 48%; semisolid; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 1.05–1.10$ (m, 3H, CH_3), 2.44–2.51 (m, 2H, CH_2CH_3), 4.11 (d, $J = 5.2$ Hz, 2H, $PhOCH_2$), 4.13 (s, 2H, $PhCH_2$), 4.50 (d, $J = 5.3$ Hz, 2H, OCH_2CH), 5.14 (s, 2H, NCH_2O), 5.16–5.32 (m, 4H, $2CH_2{=}CH$), 5.81–6.07 (m, 2H, $2CH{=}CH_2$), 6.80–7.27 (m, 4H, H_{arom}), 9.73 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.73$ (CH_3), 19.10 (CH_2CH_3), 33.34 (CH_2Ph), 68.74 ($PhOCH_2$), 70.47 ($2OCH_2CH$), 72.48 (NCH_2O), 112.99 (C-5), 117.65, 117.86 ($2CH_2{=}CH$), 114.11, 116.97, 119.76, 130.17, 136.66, 148.80 (C_{arom}), 132.89, 133.57 ($2CH{=}CH_2$), 151.92 (C-6) 159.20, 163.33 (2CO) ppm; HRMS (MALDI, peak matching): $m/z = 379.1623$ ($M + Na^+$) calcd 379.1628.

6-[3-(But-3-enyl)benzyl]-1-allyloxymethyl-5-ethyl-1H-pyrimidin-2,4-dione (**11b**, $C_{21}H_{26}N_2O$)

Yield 67%; semisolid; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 1.04–1.98$ (m, 3H, CH_3), 2.30–2.35 (m, 2H, CH_2CH_3), 2.47–2.52 (m, 2H, $CH_2CH{=}CH_2$), 2.65–2.71 (m, 2H, $PhCH_2CH_2$), 4.11 (s, 2H, CH_2Ph), 4.20 (s, 2H, NCH_2O), 5.03–5.16 (m, 2H, OCH_2CH), 5.26–5.33 (m, 4H, $2CH_2{=}CH$), 5.80–5.91 (m, 2H, $2CH{=}CH_2$), 6.92–7.27 (m, 4H, H_{arom}), 10.01 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.72$ (CH_3), 19.09 (CH_2CH_3), 33.37 ($PhCH_2CH_2$), 35.10 (CH_2Ph), 35.30 ($CH_2CH{=}CH_2$), 70.47 (OCH_2), 72.68 (NCH_2O), 115.13 (C-5), 116.91, 117.62 ($2CH_2$ =CH), 124.75, 127.34, 127.38, 129.09, 137.59, 142.94 (C_{arom}), 133.59, 135.01 ($2CH{=}CH_2$), 149.09 (C-6) 152.07, 163.53 (2CO) ppm; HRMS (MALDI, peak matching): $m/z = 377.1825$ ($M + Na^+$) calcd 377.1836.

*General Procedure for Synthesis of the Aldehydes **12a** and **12b***

Compound **9a** or **9b** (0.75 mmol) was dissolved in 15 cm^3 dry CH_2Cl_2 and cooled to −78°C. O_3 was bubbled through the solution until a blue colour appeared (*ca.* 2.5 min). Then O_2 was bubbled through the solution until the blue colour disappeared. Dimethyl sulfide (0.28 g, 4.5 mmol) was then added at −78°C, and the solution was allowed to warm to room temperature and stirred overnight. Then the solvent was evaporated under reduced pressure and the residue was chromatographed using methylene chloride/ethyl acetate (1/1, *v*/*v*) to afford compounds **12a** and **12b**.

[3-(3-Ethoxymethyl-5-ethyl-2,6-dioxo-1,2,3,6-tetrahydropyrimidin-4-ylmethyl)phenoxy]acetaldehyde (**12a**, $C_{18}H_{22}N_2O_5$)

Yield 28%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 1.04–1.25$, (m, 6H, $2CH_3$), 2.45–2.50 (m, 2H, CH_2CH_3), 3.37 (d, $J = 2.3$ Hz, 2H, OCH_2CHO), 3.59–3.62 (m, 2H, OCH_2CH_3), 4.14 (s, 2H, CH_2Ph), 4.58 (s, 2H, NCH_2O), 6.72–7.30 (m, 4H, H_{arom}), 9.84 (s, 1H, NH) 9.85 (s, 1H, CHO) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.71$, 14.97 ($2CH_3$), 19.16 (CH_2CH_3), 33.23 (CH_2Ph), 64.97 (OCH_2CH_3), 72.62 (NCH_2O), 74.94 ($PhOCH_2$), 112.50 (C-5), 114.34, 116.96, 120.81, 130.46, 137.32, 148.80 (C_{arom}), 151.81 (C-6), 158.22, 163.24 (2CO), 198.54 (CHO) ppm; HRMS (MALDI, peak matching): $m/z = 396.1420$ ($M + Na^+$) calcd 369.1421.

3-[3-(3-Ethoxymethyl-5-ethyl-2,6-dioxo-1,2,3,6-tetrahydropyrimidin-4-ylmethyl)phenyl]propanal (**12b**, $C_{19}H_{24}N_2O_4$)

Yield 18%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 1.04–1.25$ (m, 6H, $2CH_3CH_2$), 2.45–2.48 (m, 2H, CH_2CH_3), 2.74–2.79 (m, 2H, $PhCH_2CH_2$), 2.91–2.96 (m, 2H, CH_2CHO), 3.57–3.64 (m, 2H,

OCH_2CH_3), 4.13 (s, 2H, CH_2Ph), 5.11 (s, 2H, NCH_2O), 6.94–7.28 (m, 4H, H_{arom}), 9.71 (s, 1H, NH), 9.80 (s, 1H, CHO) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.70$, 14.98 ($2CH_3$), 19.12 (CH_2CH_3), 27.87 ($PhCH_2CH_2$), 33.28 (CH_2Ph), 45.06 (CH_2CHO), 64.94 (OCH_2), 72.67 (NCH_2O), 118.88 (C-5), 125.15, 127.20, 127.31, 129.40, 135.55, 141.49 (C_{arom}), 149.01 (C-6), 151.94, 163.40 (2CO), 201.04 (CHO) ppm; HRMS (MALDI, peak matching): $m/z = 367.1627$ ($M + Na^+$) calcd 367.1628.

[3-(3-Ethoxymethyl-5-ethyl-2,6-dioxo-1,2,3,6-tetrahydropyrimidin-4-ylmethyl)phenoxy]acetaldehyde oxime (**13a**)

Compound **12a** (0.34 g, 1 mmol) was dissolved in 50 cm^3 absolute ethanol. Hydroxylamine hydrochloride (0.06 g, 1 mmol) and triethylamine (0.20 g, 2 mmol) were added. The reaction mixture was refluxed for 2 h. The ethanol was removed *in vacuo* and the residue was chromatographed using methylene chloride/ethyl acetate (1/1, *v*/*v*) to afforded compound **13a**.

Yield 38%; oil; ^1H NMR (300 MHz, $CDCl_3$): $\delta = 1.05$–1.25, (m, 6H, $2CH_3$), 2.46–2.48 (m, 2H, CH_2CH_3), 3.59 (m, 2H, OCH_2CH), 3.62–3.63 (m, 2H, OCH_2CH_3), 4.13 (s, 2H, CH_2Ph), 4.78 (s, 2H, NCH_2O), 4.88–5.10 (m, 1H, CH=NOH), 6.68–7.57 (m, 4H, H_{arom}), 9.74 (br.s, 1H, OH) 10.14 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.61$,14.90 ($2CH_3$), 19.01 (CH_2CH_3), 33.23 (CH_2Ph), 61.75 ($PhOCH_2$), 64.76 (OCH_2CH_3), 72.62 (NCH_2O), 112.95 (C-5), 113.48, 116.91, 120.33, 130.32, 136.88, 148.78 (C_{arom}), 146.52 (C=NOH), 152.00 (C-6), 158.56, 163.77 (2CO) ppm.

*General Procedure for Synthesis of the Epoxide **14a** and **14b***

Compound **9a** or **9b** (1.25 mmol) was dissolved in 20 cm^3 dry $CHCl_3$ at room temperature. *MCPBA* (235 mg, 1.5 mmol) was added and the reaction mixture was stirred at room temperature under nitrogen for 3 days. Then 20 cm^3 $CHCl_3$ were added and the organic phase was washed with 2×20 cm^3 sat aqueous $Na_2S_2O_3$, 2×20 cm^3, sat aqueous $NaHCO_3$, and 2×20 cm^3 brine. The organic phase was dried ($MgSO_4$), and evaporated under reduced pressure. The products **14a** and **14b** were isolated after column chromatography (ethyl acetate/petroleum ether (60–80°C) (1/1, *v*/*v*).

6-[-3-(2-Oxiranylmethoxy)benzyl]-1-ethoxymethyl-5-ethyl-1H-pyrimidine-2,4-dione (**14a**, $C_{19}H_{24}N_2O_5$)

Yield 18%; oil; ^1H NMR (300 MHz, $CDCl_3$): $\delta = 0.85$–0.91, 1.06–1.09 (2m, 6H, $2CH_3CH_2$), 2.45–2.48 (m, 2H, CH_2CH_3), 2.74, (m, 2H, CH_2 epoxid), 3.34–3.37 (m, 2H, OCH_2CH), 3.60–3.62 (m, 2H, OCH_2CH_3), 3.92–3.94 (m, 1H, CH epoxide), 4.12 (s, 2H, CH_2Ph), 5.11 (s, 2H, NCH_2O), 6.69–7.27 (m, 4H, H_{arom}), 9.88 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.68$, 14.95 ($2CH_3$), 19.05 (CH_2CH_3), 33.22 (CH_2Ph), 44.51 (CH_2 epoxide), 49.95 (CH epoxide), 64.89 (OCH_2CH_3), 68.65 ($PhOCH_2$), 72.62 (NCH_2O), 112.73 (C-5), 114.04, 116.85, 120.09, 130.17, 136.87, 148.80 (C_{arom}), 151.93 (C-6), 159.03, 163.42 (2CO) ppm; HRMS (MALDI, peak matching): $m/z = 383.1567$ ($M + Na^+$) calcd 383.1577.

6-[-3-(2-Oxiranylethyl)benzyl]-1-ethoxymethyl-5-ethyl-1H-pyrimidine-2,4-dione (**14b**, $C_{20}H_{26}N_2O_4$)

Yield 37%; oil; ^1H NMR (300 MHz, $CDCl_3$): $\delta = 1.04$–1.20 (m, 6H, $2CH_3CH_2$), 1.78–1.86 (m, 2H, CH_2CH_3), 2.46–2.48 (m, 2H, $PhCH_2CH_2$), 2.74–2.80 (m, 4H, $PhCH_2CH_2$, CH_2 epoxid), 2.93–2.95 (m, 1H, CH epoxide), 3.57–3.64 (m, 2H, OCH_2CH_3), 4.13 (s, 2H, CH_2Ph), 5.11 (s, 2H, NCH_2O), 6.95–7.27 (m, 4H, H_{arom}), 9.55 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.72$, 15.00 ($2CH_3$), 19.14 (CH_2CH_3), 32.11 ($PhCH_2CH_2$), 33.32 (CH_2Ph), 34.14 (CH_2CH), 47.11 (CH_2 epoxid), 51.64 (CH epoxide), 64.95 (OCH_2), 72.68 (NCH_2O), 116.82 (C-5), 124.93, 127.30, 129.27, 135.37, 142.41 (C_{arom}), 149.13 (C-6) 151.90, 163.34 (2CO) ppm; HRMS (MALDI, peak matching): $m/z = 381.1773$ ($M + Na^+$) calcd 381.1785.

6-[3-(Allyloxy)benzyl]-2-methylthio-5-ethyl-1H-pyrimidine-4-one (**15**, $C_{17}H_{20}N_2O_2S$)
Compound **7a** (0.6 g, 2 mmol) was dissolved in 30 cm^3 dry *DMF* under nitrogen and methyl iodide (0.56 g, 4 mmol, 0.26 cm^3) was added. The reaction mixture was stirred at room temperature for 48 h and the solvent was evaporated under reduced pressure. The residue was chromatographed using ethyl acetate/petroleum ether (60–80°C) (1/1, *v*/*v*) to give **15**.

Yield 35%; semisolid; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 1.05$–1.11 (m, 3H, CH_3CH$_2$), 2.50 (s, 3H, SCH_3), 2.53–2.58 (m, 2H, CH_2CH$_3$), 3.87 (s, 2H, CH_2Ph), 4.49 (d, 2H, $J = 5.2$ Hz, OCH_2), 5.28–5.35 (m, 2H, CH_2=CH), 6.00–6.08 (m, 1H, CH=CH$_2$), 6.74–7.20 (m, 4H, H_{arom}), 12.64, (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.15$ (CH_3), 13.19 (SCH_3), 18.76 (*C*H$_2$CH$_3$), 40.46 (*C*H$_2$Ph), 68.69 (PhO*C*H$_2$), 112.38 (C-5), 117.61 (*C*H$_2$=CH), 133.27 (*C*H=CH$_2$), 115.62, 121.52, 122.17 129.20, 139.80, 157.03 (C_{arom}), 158.56 (C-6), 161.46 (CO), 165.08 (C=N); HRMS (MALDI, peak matching): $m/z = 339.1127$ ($M + Na^+$) calcd 339.1138.

*General Procedure for Synthesis of Compounds **16a** and **16b***
Compound **7a** (0.61 g, 2.03 mmol) was dissolved in 20 cm^3 dry *DMF*. K_2CO_3 (0.27 g, 1.95 mmol) and isopropyl bromide or isobutyl bromide (2.33 mmol) were added. The reaction mixture was stirred at room temperature for 24 h, then poured on cold H_2O and extracted with ethyl acetate. The organic phase was washed with 2×50 cm^3 sat aqueous NaCl, dried over $MgSO_4$, and evaporated under reduced pressure. The products **16a** and **16b** were isolated after column chromatography (chloroform/petroleum ether (60–80°C) (70/30, *v*/*v*).

4-[3-(Allyloxy)benzyl]-5-ethyl-6-isopropoxy-2-(isopropylsulfanyl)pyrimidin (**16a**, $C_{22}H_{30}N_2O_2S$)
Yield 72%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 0.93$–0.98 (m, 3H, CH_3CH$_2$), 1.31 (d, $J = 7.3$ Hz, 6H, 2CH$_3$), 1.41 (d, $J = 7.1$ Hz, 6H, 2CH$_3$), 2.46–2.53 (m, 2H, CH_2CH$_3$), 3.88–3.90 (m, 1H, $SCH_{Pr}i$), 3.96 (s, 2H, CH_2Ph), 4.49 (d, $J = 1.3$ Hz, 2H, PhOCH_2), 5.23–541 (m, 3H, CH_2=CH, $OCH_{Pr}i$), 5.97–6.07 (m, 1H, CH=CH$_2$), 6.72–7.25 (m, 4H, H_{arom}) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.19$ (CH_3 at C-5), 18.27 (*C*H$_2$CH$_3$), 21.91, 23.07 (2s, 4$CH_{3Pr}i$), 35.60 (SCH), 40.53 (*C*H$_2$Ph), 68.69 (PhO*C*H$_2$), 69.04 ($OCH_{Pr}i$), 112.33 (C-5), 117.60 (*C*H$_2$=CH), 133.29 (*C*H=CH$_2$), 115.47, 116.69, 121.42, 129.18, 131.19, 140.15 (C_{arom}), 158.56 (C-6), 165.33, (CO), 167.11 (C=N) ppm; HRMS (MALDI, peak matching): $m/z = 409.1923$ ($M + Na^+$) calcd 409.1920.

4-[3-(Allyloxy)benzyl]-5-ethyl-6-isobutyloxy-2-(isobutylsulfanyl)pyrimidin (**16b**, $C_{24}H_{34}N_2O_2S$)
Yield 68%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 0.94$–0.99 (m, 3H, CH_3CH$_2$), 1.01 (d, $J = 3.6$ Hz, 6H, 2CH$_3$), 1.02 (d, $J = 6.7$ Hz, 6H, 2CH$_3$), 2.01–2.09 (m, 2H, 2$CH_{But}i$), 2.51–2.59 (m, 2H, CH_2CH$_3$), 2.98 (d, $J = 6.7$ Hz, 2H, SCH_2), 3.97 (s, 2H, CH_2Ph), 4.09 (d, $J = 6.4$ Hz, 2H, OCH_2) 4.47 (d, $J = 5.2$ Hz, 2H, PhOCH_2), 5.23–5.24 (m, 2H, CH_2=CH), 5.99–6.07 (m, 1H, CH=CH$_2$), 6.72–7.25 (m, 4H, H_{arom}), 12.64 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 13.29$ (CH_3 at C-5), 18.29 (*C*H$_2$CH$_3$), 19.21, 21.95 (4$CH_{3But}i$), 27.86, 28.68 ($CH_{But}i$), 39.43 (SCH_2), 40.48 (*C*H$_2$Ph), 68.69 (PhO*C*H$_2$), 72.59 ($OCH_{2But}i$), 112.37 (C-5), 117.61 (*C*H$_2$=CH), 133.28 (*C*H=CH$_2$), 115.43, 116.49, 121.42, 129.20, 131.18, 140.19, (C_{arom}), 158.59 (C-6), 165.25, (CO), 167.48 (C=N) ppm; HRMS (MALDI, peak matching): $m/z = 437.2224$ ($M + Na^+$) calcd 437.2233.

6-[3-(Allyloxy)benzyl]-2-dimethylamino-5-ethyl-1H-pyrimidine-4-one (**17**, $C_{18}H_{23}N_3O_2$)
The β-ketoester **5a** was dissolved in *Et*ONa (Na, 0.14 g, 6.3 mmol in 50 cm^3 abs *Et*OH). *N*,*N*-Dimethylguanidine sulfate (1.17 g, 4.3 mmol) was added and the reaction mixture was refluxed for 48 h, cooled, filtered, evaporated to the half volume *in vacuo*, poured into H_2O, and extracted with chloroform. The organic layer was washed with H_2O (2×50 cm^3), dried over $MgSO_4$, and evaporated *in vacuo*. The residue was chromatographed using $CHCl_3$/*Me*OH (9/1, *v*/*v*). Yield 38%; oil; ^{1}H NMR (300 MHz, $CDCl_3$): $\delta = 0.96$–1.01 (m, 3H, CH_3), 2.42–2.49 (m, 2H, CH_2CH$_3$), 3.11 (s, 6H, 2NCH_3), 3.78 (s, 2H, PhCH_2), 4.49 (d, $J = 5.3$ Hz, 2H, OCH_2CH), 5.23–5.27 (m, 2H, CH_2 = CH), 6.01–6.08 (m, 1H, CH = CH_2), 6.72–7.25 (m, 4H, H_{arom}), 9.74 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$):

$\delta = 13.72$ (CH_3), 18.44 (CH_2CH_3), 37.20 ($2NCH_3$), 40.91 (CH_2Ph), 68.65 ($PhOCH_2$), 112.13 (C-5), 117.52 ($CH_2 = CH$), 133.35 ($CH{=}CH_2$), 112.72, 115.50, 121.51, 129.01, 140.61, 152.03 (C_{arom}), 158.46 (C-6), 163.44 (CO), 165.94 (NC=N) ppm; HRMS (MALDI, peak matching): $m/z = 314.2001$ ($M + H^+$) calcd 314.1980.

Antiviral Assay Procedures

Compounds were solubilized in *DMSO* at 200 mM and then diluted in culture medium.

Cells and Viruses

MT-4, C8166, and H9/IIIB cells were grown at 37°C in a 5% CO_2 atmosphere in RPMI 1640 medium supplemented with 10% fetal calf serum (FCS), 100 IU/cm^3 penicillin G, and 100 μg/cm^3 streptomycin. Cell cultures were checked periodically for the absence of mycoplasma contamination with a MycoTect Kit (Gibco). Human immunodeficiency viruses type 1 (HIV-1, IIIB strain) were obtained from supernatants of persistently infected H9/IIIB cells. The HIV-1 stock solutions had titers of 4.5×10^6 50% cell culture infectious dose ($CCID_{50}$)/cm^3. The K103R-V179D-P225H mutant was derived from a IIIB strain passaged in C8166 cells in the presence of efavirenz (up to 2 μM). The Y181C mutant (NIH N119) was derived from an *AZT*-sensitive clinical isolate passaged initially in CEM and then in MT-4 cells in the presence of nevirapine (10 μM). The K103N + Y181C (NIH A17) was derived from the IIIB strain passaged in H9 cells in the presence of BI-RG 587 (1 μM). K103R + V179D + P225H (EFV^R), Y181C, and K103N + Y181C stock solutions had titers of 3.0×10^5, 1.3×10^6, and 2.5×10^5 $CCID_{50}$/cm^3, respectively.

HIV Titration

Titration of HIV was performed in C8166 cells by the standard limiting dilution method (dilution 1:2, four replica wells per dilution) in 96-well plates. The infectious virus titer was determined by light microscope scoring of syncytia after 4 days of incubation. Virus titers were expressed as $CCID_{50}$/cm^3.

Anti-HIV Assays

The activity of test compounds against multiplication of wild type HIV-1, Y181C, and K103N + Y181C in acutely infected cells was based on inhibition of virus-induced cytopathicity in MT-4 cells. The activity of the compounds against the EFV^R multiplication in acutely infected cells was based on inhibition of p24 antigen in C8166 cells. Briefly, an amount of 50 mm^3 of culture medium containing 1×10^4 cells was added to each well of flat-bottom microtiter trays containing 50 mm^3 of culture medium with or without various concentrations of test compounds. Then an amount of 20 mm^3 of HIV suspensions (containing the appropriate amount of $CCID_{50}$ to cause complete cytopathicity at day 4) was added. After incubation at 37°C, cell viability was determined by the 3-(4,5-dimethylthiazol-1-yl)-2,5-diphenyltetrazolium bromide (*MTT*) method [16]. Alternatively, p24 levels were determined by an immunoenzymatic kit (Abbott). The cytotoxicity of test compounds was evaluated in parallel with their antiviral activity and was based on the viability of mock-infected cells, as monitored by the *MTT* method.

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