Monatshefte für Chemie 133, 723–734 (2002)

Monatshefte für Chemie
Chemical Monthly

Anti-HIV Active Naphthyl Analogues of *HEPT* and *DABO*

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Summary. 5-Isopropyl-6-naphthyl uracil and 5-isopropyl-6-naphthyl-2-thiouracil were alkylated to give N-1-(ethoxymethyl and methylthiomethyl) uracil and S^2-cyclohexyl-thiouracil, respectively. 5-Ethyl-6-naphthyl uracil and 5-ethyl-6-naphthyl-2-thiouracil afforded N-1-(ethoxymethyl, methoxymethyl, methylthiomethyl, acetoxyethoxy methyl and hydroxyethoxy methyl) uracil and S^2-((2,2-diethoxyethyl), methoxycarbonylmethyl, ethoxycarbonylpropyl, methylthiomethyl, ethoxymethyl, methyl and cyclohexyl)-thiouracil upon alkylation.

Keywords. Non-nucleoside reverse transcriptase inhibitors; HIV-1; MKC-442 analogues; S-*DABO*; Uracil-6-naphthyl analogues.

Introduction

Reverse transcriptase (RT), being the pivot in the human immunodeficiency virus type 1 (HIV-1) replication, is still one of the most attractive targets for the development of new antiretroviral agents. Among the non-nucleoside inhibitors of RT, 1-((2-hydroxyethoxy)-methyl)-6-(phenylthio)-thymine (*HEPT*) has been considered an interesting compound for the synthesis of new derivatives with activity against HIV [1, 2], *e.g.* MKC-442 [3]. Various thio analogues of dihydroalkoxy benzyloxopyrimidines (S-*DABO*s) have also been found to inhibit HIV-1 [4].

Correct spatial positioning of the phenyl group in MKC-442 seems to be a prerequisite of its activity against HIV-1. The conformation of MKC-442 in a complex with HIV reverse transcriptase enzyme has been determined by X-ray crystallography [5]. It has been suggested that a major determinant of the increased potency of MKC-442 is an improved interaction between residue *Tyr*181 in the protein and the 6-benzyl ring of the inhibitor which stabilizes the structure of the complex. There are numerous examples of 6-aroyl analogues **1** claimed to have potent activity against HIV-1 [6]. However, loss of activity against HIV when

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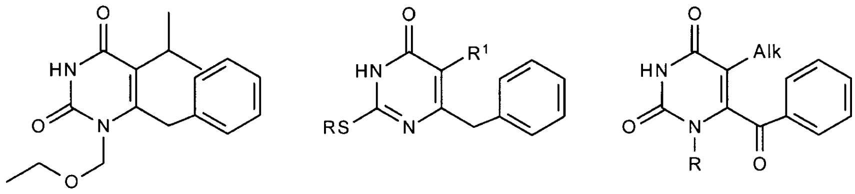


Scheme 1

compared with MKC-442 is found for numerous compounds with the phenyl group locked into other conformations than that determined by X-ray for MKC-442 or with the phenyl group in regioisomeric positions [7]. Strange as it may seem there has only been little interest in the 6-aryl analogues of MKC-442 [8], but this could be a consequence of the former observation of the importance of proper placing the phenyl group.

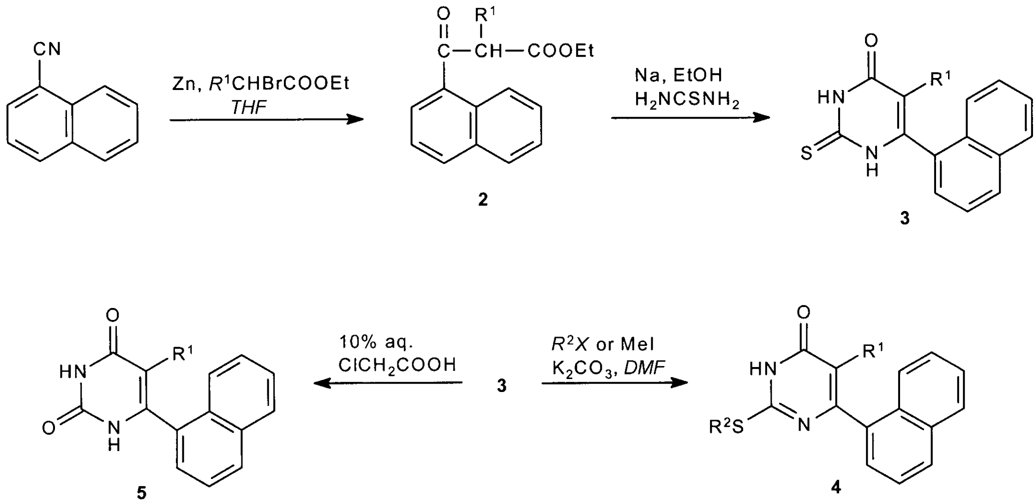
In the structure of the 6-aroyl derivatives **1** it is likely that the carbonyl group and the aryl group are in the same plane perpendicular to the uracil ring due to substituents in its 1- and 5-position. In this way, the aryl group is positioned in a similar way as in MKC-442 and capable of interaction with *Tyr*181 in the reverse transciptase enzyme. In the present work, the planar 6-aroyl group in **1** is replaced with a 1-naphtyl group, assuming that the second ring of the naphthalene ring is regioidentical to the aryl group of **1** and that this could result in new active compounds against HIV-1.

Results and Discussion

Chemistry

Ethyl 2-alkyl-3-naphthyl-3-oxopropionates **2a**,**b** were obtained from commercially available 1-cyanonaphthalene by a method previosly described [9]. The β-ketoesters **2a**,**b** were condensed by the method of *Danel et al.* [2a] with thiourea in the presence of sodium ethoxide to furnish the corresponding 5-alkyl-6-naphthyl-2-thiouracils **3a**,**b**, which in turn in a standard reaction [10] underwent exchange of sulfur with oxygen by boiling in aqueous chloroacetic acid to afford the corresponding uracils **5a**,**b**. In this reaction sequence the impure raw materials from the synthesis of **2a**,**b** were used without further purification for the synthesis of **3a**,**b**. The NMR spectra of crude **2a**,**b** showed an impurity believed to be a β-ketoester resulting from self-condensation of alkyl 2-bromobutyrate. Upon reaction with thiourea this β-ketoester impurity also formed a uracil derivative as an impurity in the raw compounds **3a**,**b**. Pure **3a**,**b** were obtained by column chromatography.

Reaction of 5-ethyl-6-naphthyl-2-thiouracil (**3a**) with bromoacetaldehyde diethylacetal, methyl bromoactate, ethyl 4-bromobutyrate, methylthiomethyl chloride, ethoxymethyl chloride, methyl iodide, or cyclohexyl chloride in *DMF* in the presence of potassium carbonate and likewise reaction of 5-isopropyl-6-naphthyl-2-thiouracil (**3b**) with cyclohexyl chloride afforded 2-alkylthio-6-naphthylpyrimidin-4(1*H*)-ones **4a**–**g** and **4h**, respectively.



2,3,5	*R*[1]
a	Et
b	*i*-Pr

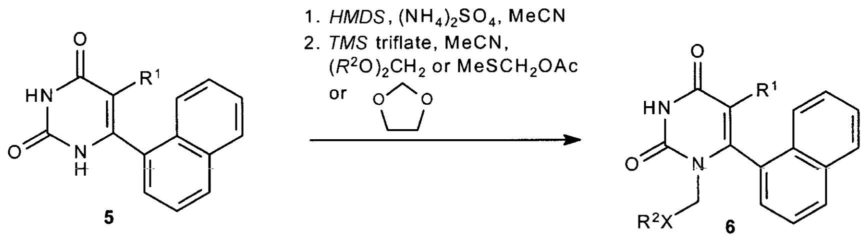
4	*R*[1]	*R*[2]
a	Et	$CH_2CH(OEt)_2$
b	Et	CH_2COOMe
c	Et	$(CH_2)_3COOEt$
d	Et	CH_2SCH_3
e	Et	CH_2OEt
f	Et	Me
g	Et	Cyclohexyl
h	*i*-Pr	Cyclohexyl

Scheme 2

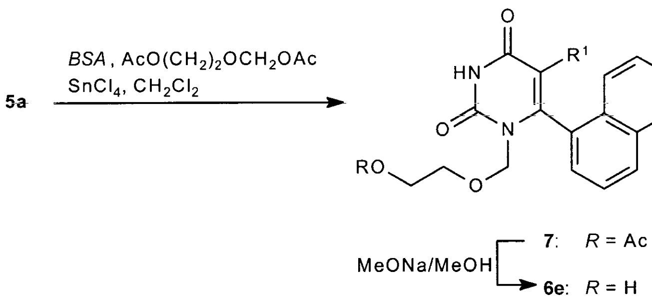
The uracil derivatives **5a**,**b** were silylated by heating at reflux in 1,1,1,3,3,3-hexamethyldisilazane (*HMDS*) and underwent alkylation by treatment with methylthiomethyl acetate, dimethoxymethane, diethoxymethane, or 1,3-dioxalane in the presence of trimethylsilyl trifluoromethanesulfonate (*TMS* triflate) according to the method of *Vorbrüggen et al.* [11] to afford the acyclic nucleosides **6a**–**f**.

In an alternative procedure for the synthesis of **6e**, uracil **5a** was treated with N,O-*bis*-trimethylsilylacetamide (*BSA*) in methylene chloride. Subsequent addition of acetoxyethyl acetoxymethyl ether and tin(IV) chloride at 0°C afforded the acyclic nucleoside 1-((2-acetoxyethoxy)-methyl)-5-ethyl-6-(naphthyl)-uracil (**7**) which was deprotected by sodium methoxide in methanol to afford **6e**.

The naphthyl group in the acyclic nucleosides **6** was, as expected, found to be oriented perpendicular to the uracil ring and induced chirality in the molecules. This was confirmed by ^{1}H NMR spectra which showed large shift differences (0.5–1.0 ppm) for the two diasterotopic protons of the methylene group at N-1 due to the shielding effect of the naphthyl ring, thus pointing to N-1 alkylation. The N-1 alkylation was also corroborated by a strong NOE at the aromatic protons when each of the protons in the methylene group at N-1 of **6a** were irradiated. Even for compounds **3**–**5** without N-1 substituents, the naphthyl ring was locked in a vertical position compared to the uracil ring. This was deduced by observing shift differences in the ^{1}H NMR spectra for the two diasterotopic protons in the ethyl group at C-5 of the uracil ring.



6	R^1	R^2X
a	Et	MeO
b	Et	EtO
c	*i*-Pr	EtO
d	*i*-Pr	MeS
e	Et	$HO(CH_2)_2O$
f	Et	MeS



Scheme 3

Antiviral activity

The target compounds **6a–f** (Table 1) indeed showed activity against HIV-1 when tested as described earlier [2b]. This is the first time to find activity against HIV-1 for an MKC-442 analogue with an aryl group connected directly at C-6 in the uracil ring, the trick being to use a polyaromatic ring as the substituent which can place an aromatic ring in nearly the same position as the phenyl ring of MKC-442. This assumption is likely as chirality could be deduced for compounds **6** from their NMR spectra which originates from an out-of-plane conformation of the naphthyl ring. The activity of compounds **6** is very sensitive to the type of N-1 substituent. In fact, only an ethoxymethyl group at N-1 results in a significant activity against HIV-1. The more bulky isopropyl group at C-5 improved the activity 10 times compared with the corresponding compound with an ethyl group. Although the most active compound **6c** showed $ED_{50} = 0.4\,\mu M$, its activity is still *ca.* 100 times lower than that of MKC-442 and 10 times lower than that of the homologue of **6c** where a methylene group is inserted between the naphthyl ring and the uracil ring [2c]. The corresponding 6-naphthyl S-*DABO* derivatives **4a–h** were also synthesized and investigated for their activity against HIV-1, but only moderate activities were found. Hoping to find a new generation of MKC-442 analogues with a resistance

Table 1. Antiviral activity against HIV-1 in MT-4 cells

	ED_{50} (μM)[a]	CD_{50} (μM)[b]
4a	–[c]	28
4b	32	>100
4c	37	>100
4d	–[c]	32
4e	27	>100
4f	37	63
4g	63	100
4h	>100	>100
6a	25	>100
6b	4.0	>100
6c	0.4	>100
6d	16	100
6e	18	>100
6f	32	>100
7	18	>100
MKC-442	0.005	141

[a] Effective dose of compound achieving 50% inhibition of HIV-1 antigen production in MT-4 cultures; [b] cytoxic dose of compound required to reduce proliferation of normal uninfected MT-4 cells by 50%; [c] not active at subtoxic concentration

profile differing from that of the existing MKC-442 analogues, compounds **4** and **6** were also tested against the N119 (*Tyr*181*Cys*) mutant strain; however, no activity was found.

Experimental

NMR spectra were recorded on a Bruker AC-250 FT NMR spectrometer at 250 MHz for ^{1}H and 62.9 MHz for ^{13}C with *TMS* as an internal standard. Analytical silica gel TLC plates 60 F_{254} and the silica gel (0.040–0.063) used for column chromatography were purchased from Merck. *THF* was distilled from sodium benzophenone prior to use.

Typical procedure for the preparation of **2a**,**b**

Activated zinc dust (zinc dust washed sequentially with 3 *M* HCl, dist. H_2O, EtOH, and dry Et_2O and then dried *in vacuo*, 45 g, 0.69 mol), was suspended in refluxing *THF* (400 cm^3) under nitrogen. A few drops of alkyl-2-bromobutyrate were added to initiate the reaction. After the appearance of a green colour (approx. 60 min), 1-cyano naphthylene (0.14 mol) was added in one portion followed by slow addition of alkyl-2-bromobutyrate (0.36 mol). The mixture was refluxed for additional 20 min. After cooling and dilution with *THF* (1240 cm^3), 50% K_2CO_3 (180 cm^3) was added, and the mixture was stirred vigorously. The *THF* layer was decanted, and the residue was extracted with *THF* (3×100 cm^3). The combined organic fractions were treated with 10% aq. HCl (150 cm^3) at room temperature for 45 min. The mixture was concentrated *in vacuo*. CH_2Cl_2 was added, and the solution was washed with sat. $NaHCO_3$, dried over Na_2SO_4, and evaporated under reduced pressure to furnish the crude oxoester **2a**,**b** which was used without further purification. Analytically pure **2a**,**b** was obtained by preparative TLC (10% EtOAc in petroleum ether).

Ethyl 2-ethyl-3-(naphth-1-yl)-3-oxobutyrate (**2a**; $C_{17}H_{18}O_3$)

^{1}H NMR ($CDCl_3$, δ, 250 MHz): 0.88–0.95 (m, 6H, 2 × CH_3), 1.90–1.92 (m, 2H, CH_2), 3.94–3.97 (m, 3H, CH, CH_2), 7.58–8.30 (m, 7H, H-arom.) ppm; ^{13}C NMR ($CDCl_3$, δ, 75 MHz): 11.43 (CH_3), 13.59 (CH_3), 21.65 (CH_2), 57.63 (CH), 60.52 (CH_2), 124.75, 125.10, 126.62, 127.98, 128.09, 128.56, 129.59, 132.84, 133.51, 135.19 (C-arom.), 168.73 (COOEt), 197.25 (C=O) ppm.

Ethyl 2-isopropyl-3-(naphth-1-yl)-3-oxobutyrate (**2b**; $C_{18}H_{20}O_3$)

^{1}H NMR ($CDCl_3$, δ, 250 MHz): 0.95–1.12 (m, 9H, 3 × CH_3), 2.48–2.53 (m, 1H, CH), 4.12–4.14 (q, 2H, J = 7.0 Hz, CH_2), 4.50 (m, 1H, CH), 7.58–8.30 (m, 7H, H-arom.) ppm; ^{13}C NMR ($CDCl_3$, δ, 75 MHz): 18.97 (CH_3), 20.78 (2 × CH_3), 29.10 (CH), 62.02 (CH_2), 63.17 (CH), 125.53, 127.12, 127.22, 127.50, 128.02, 128.14, 131.42, 136.19, 148.29 (C-arom.), 168.73 (COOEt), 197.25 (C=O) ppm.

Typical procedure for the preparation of **3**

Crude compounds **2a**,**b** (0.14 mol) was added to a solution of Na (9.3 g, 0.40 mol) and thiourea (21.3 g, 0.28 mol) in EtOH (200 cm^3). The mixture was heated at reflux overnight. After cooling, the solvent was removed *in vacuo*, and the residue was dissolved in H_2O and neutralized with HCl. The precipitate was collected, washed with H_2O, and recrystallized from EtOH to give **3** as white crystals.

5-Ethyl-6-(naphth-1-yl)-2-thiouracil (**3a**; $C_{16}H_{14}N_2OS$)

Yield: 10 g (20%); m.p.: 192–195°C; MS (EI): m/z = 282 (M^+); ^{1}H NMR (*DMSO*-d_6, δ, 250 MHz): 0.77 (t, 3H, J = 7.3 Hz, CH_3), 1.79, 2.01 (2 × m, 2H, CH_2), 7.55–8.01 (m, 7H, H-arom.), 12.43, 12.58 (2 × s, 2H, 2 × NH) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 13.01 (CH_3), 18.67 (CH_2), 118.47 (C-5), 124.61, 125.44, 126.56, 126.94, 127.28, 128.54, 129.38, 129.95, 130.26, 133.04 (C-arom.), 148.20 (C-6), 161.58 (C-4), 174.54 (C-2) ppm.

5-Isopropyl-6-(naphth-1-yl)-2-thiouracil (**3b**; $C_{17}H_{16}N_2OS$)

Yield: 10 g (20%); m.p.: 303–305°C; MS (EI): m/z = 296 (M^+); ^{1}H NMR (*DMSO*-d_6, δ, 250 MHz): 0.99, 1.01 (2 × d, 6H, J = 6.9 Hz, 2 × CH_3), 2.08 (hept, 1H, J = 6.9 Hz, CH), 7.49–8.05 (m, 7H, H-arom.), 12.32, 12.45 (2 × s, 2H, 2 × NH) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 19.57, 19.74 (2 × CH_3), 27.91 (CH), 121.11 (C-5), 124.62, 125.53, 126.64, 126.71, 127.30, 128.59, 129.86, 129.97, 130.40, 133.06 (C-arom.), 147.90 (C-6), 160.92 (C-4), 174.38 (C-2) ppm.

General procedure for the preparation of 2-(alkylthio)-5-alkyl-6-(naphth-1-yl)-pyrimidine-4(3H)-ones **4a–h**

A mixture of **3** (0.282 g, 1 mmol), alkyl halogenide (bromoacetaldehyde diethylacetal, methyl bromoactate, ethyl 4-bromobutyrate, methylthiomethyl chloride, ethoxymethyl chloride, methyl iodide, or cyclohexyl chloride; 1 mmol) and K_2CO_3 (138 mg, 1 mmol) in anhydrous *DMF* (5 cm^3) was stirred overnight at room temperature. After treatment with H_2O (100 cm^3) the solution was extracted with EtOAc (3 × 50 cm^3). The combined extracts were washed with sat. NaCl (2 × 50 cm^3), dried ($MgSO_4$), filtered, and concentrated *in vacuo* to give the crude products **4a–g** which were purified by column chromatography ($CHCl_3$).

2-((2,2-Diethoxyethyl)-thio)-5-ethyl-6-(naphth-1-yl)-pyrimidine-4(3H)-one (**4a**; $C_{22}H_{26}N_2O_3S$)

Yield: 0.25 g (63%); m.p.: 90°C; MS (EI): $m/z = 398$ (M^+); ^1H NMR (*DMSO*-d_6, δ, 250 MHz): 0.83–1.12 (m, 9H, 3 × CH_3), 1.98, 2.20 (2 × m, 2H, CH_2), 3.04–3.52 (m, 6H, 3 × CH_2), 4.55 (t, 1H, $J = 5.3$ Hz, CH), 7.43–8.05 (m, 7H, H-arom.) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 13.22 (CH_3), 14.88 (2 × CH_3), 19.48, 32.86, 61.63, 61.73 (CH_2), 101.12 (CH), 121.92, 125.29, 125.50, 126.02, 126.29, 128.18, 128.21, 130.53, 133.21, 136.72 (C-arom.), 158.86 (C-6), 159.52 (C-4), 165.88 (C-2) ppm.

5-Ethyl-2-((methoxycarbonylmethyl)-thio)-6-(naphth-1-yl)-pyrimidine-4(3H)-one (**4b**; $C_{19}H_{18}N_2O_3S$)

Yield: 0.15 g (60%); m.p.: 165°C; MS (EI): $m/z = 354$ (M^+); ^1H NMR (*DMSO*-d_6, δ, 250 MHz): 0.81 (t, 3H, $J = 7.3$ Hz, CH_3), 1.93, 2.22 (2 × m, 2H, CH_2), 3.44 (s, 3H, CH_3), 3.85–3.89 (2 × d, 2H, $J = 15.8$ Hz, CH_2), 7.40–8.04 (m, 7H, H-arom.) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 12.99 (CH_3), 19.35 (CH_2), 31.84 (CH_2), 51.81 (CH_3), 122.55, 125.18, 125.28, 125.61, 126.12, 126.44, 128.26, 128.51, 130.30, 133.15, 136.00 (C-arom.), 157.76 (C-6), 159.08 (C-4), 164.29 (C-2), 169.05 (C=O) ppm.

2-((Ethoxycarbonylpropyl)-thio)-5-ethyl-6-(naphth-1-yl)-pyrimidine-4(3H)-one (**4c**; $C_{22}H_{24}N_2O_3S$)

Yield: 0.18 g (64%); m.p.: 90°C; MS (EI): $m/z = 396$ (M^+); ^1H NMR (*DMSO*-d_6, δ, 250 MHz): 0.80 (t, 3H, $J = 7.4$ Hz, CH_3), 1.11 (t, 3H, $J = 7.1$ Hz, CH_3), 1.82 (quint, 2H, $J = 7.2$ Hz, CH_2), 1.92, 2.16 (2 × m, 2H, CH_2), 2.30 (t, 2H, $J = 7.4$ Hz, CH_2), 2.96 (m, 2H, CH_2), 3.97 (q, 2H, $J = 7.1$ Hz, CH_2), 7.39–8.00 (m, 7H, H-arom.) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 13.24 (CH_3), 13.87 (CH_3), 19.52, 24.38, 28.66, 32.28 (CH_2), 59.69 (CH_2), 121.57, 125.34, 125.53, 126.02, 126.23, 128.09, 128.21, 130.59, 133.20, 136.95 (C-arom.), 158.54 (C-6), 159.66 (C-4), 166.34 (C-2), 172.48 (C=O) ppm.

5-Ethyl-2-(methylthiomethylthio)-6-(naphth-1-yl)pyrimidine-4(3H)-one (**4d**; $C_{18}H_{18}N_2OS_2$)

Yield: 0.14 g (58%); m.p.; 135°C; MS (EI): $m/z = 342$ (M^+); ^1H NMR (*DMSO*-d_6, δ, 250 MHz): 0.77 (t, 3H, $J = 7.2$ Hz, CH_3), 1.85, 2.15 (2 × m 2H, $J = 12.1$ Hz, CH_2), 4.14, 4.21 (2 × d, 2H, CH_2), 7.34–7.96 (m, 7H, H-arom.) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 13.34 (CH_3), 14.66 (CH_3), 19.63 (CH_2), 34.94 (CH_2), 121.47, 125.37, 125.60, 125.68, 126.04, 126.24, 128.19, 130.63, 133.19, 137.11, 137.72 (C-arom.), 158.82 (C-6), 160.72 (C-4), 168.04 (C-2) ppm.

2-(Ethoxymethylthio)-5-ethyl-6-(naphth-1-yl)-pyrimidine-4(3H)-one (**4e**; $C_{19}H_{20}N_2O_2S$)

Yield: 0.15 g (63%); m.p.: 120°C; MS (EI): $m/z = 340$ (M^+); ^1H NMR (*DMSO*-d_6, δ, 250 MHz): 0.80 (t, 3H, $J = 7.3$ Hz, CH_3), 1.03 (t, 3H, $J = 7.0$ Hz, CH_3), 1.93, 2.17 (2 × m, 2H, CH_2), 3.46 (m, 2H, CH_2), 5.23, 5.25 (2 × d, 2H, CH_2), 7.39–8.00 (m, 7H, H-arom.) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 13.27 (CH_3), 14.51 (CH_3), 19.59 (CH_2), 63.86 (CH_2), 70.95 (CH_2), 121.74, 125.31, 125.53, 125.59, 125.99, 126.16, 128.17, 130.62, 133.19, 137.04 (C-arom.), 158.80 (C-6), 159.77 (C-4), 167.34 (C-2) ppm.

5-Ethyl-2-(methylthio)-6-(naphth-1-yl)-pyrimidine-4(3H)-one (**4f**; $C_{17}H_{16}N_2OS$)

Yield: 0.1 g (48%); m.p.: 160°C; MS (EI): $m/z = 296$ (M^+); ^1H NMR (*DMSO*-d_6, δ, 250 MHz): 0.80 (t, 3H, $J = 7.3$ Hz, CH_3), 1.91, 2.09 (2 × m, 2H, CH_2), 2.35 (s, 3H, CH_3), 7.43–8.03 (m, 7H, H-arom.) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 12.68 (CH_3), 19.46 (CH_3), 30.59 (CH_2), 121.71, 125.20, 125.27, 125.43, 125.93, 126.24, 128.07, 130.32, 133.00, 136.52 (C-arom.), 158.63 (C-6), 159.91 (C-4), 165.20 (C-2) ppm.

2-(Cyclohexylthio)-5-ethyl-6-(naphth-1-yl)-pyrimidine-4(3H)-one (**4g**; $C_{22}H_{24}N_2OS$)

Yield: 0.16 g (62%); m.p.: 260°C; MS (EI): $m/z = 364$ (M^+); ^{1}H NMR (*DMSO*-d_6, δ, 250 MHz): 0.78 (t, 3H, $J = 7.3$ Hz, CH_3), 1.27–2.13 (m, 12H, $6 \times CH_2$), 3.60 (m, 1H, CH), 7.36–8.00 (m, 7H, H-arom.) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 13.36 (CH_3), 19.37 (CH_2), 25.16 (CH_2), 32.65 (CH_2), 33.08 (CH_2), 41.27 (CH), 120.04, 125.31, 125.47, 125.84, 126.09, 126.27, 127.55, 128.05, 130.84, 133.17, 137.99 (C-arom.), 159.10 (C-4), 160.68 (C-4), 162.61 (C-2) ppm.

2-(Cyclohexylthio)-5-isopropyl-6-(naphth-1-yl)-pyrimidine-4(3H)-one (**4h**; $C_{23}H_{26}N_2OS$)

Yield: 0.3 g (60%); m.p.: 240°C; MS (EI): $m/z = 378$ (M^+); ^{1}H NMR ($CDCl_3$, δ, 250 MHz): 1.21, 1.26 ($2 \times$ d, 6H, $J = 6.7$ Hz, $2 \times CH_3$), 1.42–2.01 (m, 10H, $5 \times CH_2$), 2.56 (m, 1H, CH), 3.80 (m, 1H, CH), 7.25–7.91 (m, 7H, H-arom.) ppm; ^{13}C NMR ($CDCl_3$, δ, 75 MHz): 19.89 ($2 \times CH_3$), 25.40, 32.31, 33.19 (CH_2), 43.90 (CH), 125.18, 125.40, 125.78, 126.08, 126.19, 127.13, 128.30, 128.58, 131.00, 133.68 (C-arom.), 157.06 (C-6), 160.52 (C-2), 164.32 (C-4) ppm.

Typical procedure for the preparation of **5**

The crude 2-thiouracil (**3**) was suspended in boiling 10% aq. chloroacetic acid, and heating was continued until disappearance of the starting material (TLC). After cooling, the precipitate was collected, washed with H_2O, and recrystallized from EtOH to afford **5**.

5-Ethyl-6-(naphth-1-yl)-uracil (**5a**; $C_{16}H_{14}N_2O_2$)

Yield: 1.3 g (87%); m.p.: 130°C; MS (EI): $m/z = 266$ (M^+); ^{1}H NMR (*DMSO*-d_6, δ, 250 MHz): 0.76 (t, 3H, $J = 7.3$ Hz, CH_3), 1.76, 1.98 ($2 \times$ m, 2H, CH_2), 7.56–8.12 (m, 7H, H-arom.), 10.94, 11.26 ($2 \times$ s, 2H, $2 \times$ NH) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 13.55 (CH_3), 18.53 (CH_2), 112.86 (C-5), 124.74, 125.50, 126.60, 127.25, 128.51, 129.70, 130.15, 130.46, 133.14 (C-arom.), 147.82 (C-6), 150.90 (C-2), 164.57 (C-4) ppm.

5-Isopropyl-6-(naphth-1-yl)-uracil (**5b**; $C_{17}H_{16}N_2O_2$)

Yield: 1.1 g (71%); m.p.: 250°C; MS (EI): $m/z = 280$ (M^+); ^{1}H NMR (*DMSO*-d_6, δ, 250 MHz): 0.99, 1.01 ($2 \times$ d, 6H, $J = 7.0$, $2 \times CH_3$), 2.05 (hept, 1H, $J = 7.0$ Hz, CH), 7.47–8.04 (m, 7H, H-arom.), 10.79, 11.07 ($2 \times$ s, 2H, $2 \times$ NH) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 19.94, 20.15 ($2 \times CH_3$), 27.60 (CH), 115.51 (C-5), 124.74, 125.56, 126.33, 127.23, 128.52, 129.57, 130.25, 131.08, 133.06 (C-arom.), 147.38 (C-6), 150.77 (C-2), 163.87 (C-4) ppm.

General procedure for the preparation of 1-(alkoxymethyl)-5-alkyl-6-(naphth-1-yl)-uracils **6a–c**

Compounds **5a**,**b** (0.84 g, 3 mmol) were silylated with 5 cm^3 of 1,1,1,3,3,3-hexamethyldisilazane (*HMDS*) in the presence of 10 mg $(NH_4)_2SO_4$. When the silylation was complete, the excess of *HMDS* was evaporated *in vacuo* to yield a translucent yellow oil which was dissolved in anhydrous MeCN (10 cm^3) and cooled to −35°C. Trimethylsilyl trifluoromethanesulfonate (*TMS*-triflate; 0.62 g, 2.79 mmol) was added in one portion, followed by dropwise addition of dialkoxymethane (3.1 g, 30 mmol). Then the mixture was stirred for 3 h at −35°C. When the reaction was finished (TLC), the mixture was quenched with ice cold saturated $NaHCO_3$ (10 cm^3) and evaporated to near dryness by coevaporation with EtOH ($2 \times 50\,cm^3$). The resulting solid was suspended in Et_2O ($200\,cm^3$), and the mixture was stirred for 1 h. After filtration, the residue was extracted with Et_2O ($100\,cm^3$), and the combined organic fractions were evaporated to furnish crude **6a–c**. Column chromatography (10–25% EtOAc in petroleum ether) gave **6a–c** as a white powder.

5-Ethyl-1-(methoxymethyl)-6-(naphth-1-yl)-uracil (**6a**; $C_{18}H_{18}N_2O_3$)

Yield: 0.75 g (71%); m.p.: 150°C; MS (EI): $m/z = 310$ (M^+); ^{1}H NMR (*DMSO*-d_6, δ, 250 MHz): 0.65 (t, 3H, $J = 7.1$ Hz, CH_3), 1.56, 1.87 ($2 \times$ m, 2H, CH_2), 2.90 (s, 3H, CH_3), 4.25, 4.83 ($2 \times$ d, 2H, $J = 10.4$ Hz, CH_2), 7.48–8.05 (m, 7H, H-arom.) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 13.30 (CH_3), 19.50 (CH_3), 55.60 (CH_2), 74.50 (CH_2), 115.75 (C-5), 124.79, 125.30, 126.70, 127.32, 127.57, 128.48, 128.89, 129.89, 130.40, 132.90 (C-arom.), 148.80 (C-6), 151.83 (C-2), 163.60 (C-4) ppm.

1-(Ethoxymethyl)-5-ethyl-6-(naphth-1-yl)-uracil (**6b**; $C_{19}H_{20}N_2O_3$)

Yield: 0.73 g (75%); m.p.: 145°C; MS (EI): $m/z = 324$ (M^+); ^{1}H NMR ($CDCl_3$, δ, 250 MHz): 0.76 (t, 3H, $J = 7.3$ Hz, CH_3), 0.87 (t, 3H, $J = 6.9$ Hz, CH_3), 1.75, 2.05 ($2 \times$ m, 2H, CH_2), 3.18 (m, 2H, CH_2), 4.38, 4.99 ($2 \times$ d, 2H, $J = 10.0$ Hz, CH_2), 7.42–7.96 (m, 7H, H-arom.) ppm; ^{13}C NMR ($CDCl_3$, δ, 75 MHz): 13.42 (CH_3), 14.60 (CH_3), 19.89 (CH_2), 64.29 (CH_2), 73.48 (CH_2), 117.46 (C-5), 124.63, 124.98, 126.75, 127.37, 127.58, 128.63, 128.70, 130.24, 130.68, 133.33 (C-arom.), 149.79 (C-6), 151.92 (C-2), 163.58 (C-4) ppm.

1-(Ethoxymethyl)-5-isopropyl-6-(naphth-1-yl)-uracil (**6c**; $C_{20}H_{22}N_2O_3$)

Yield: 0.6 g (77%); white powder; m.p.: 155°C; MS (EI): $m/z = 338$ (M^+); ^{1}H NMR (*DMSO*-d_6, δ, 250 MHz): 0.75 (t, 3H, $J = 6.9$ Hz, CH_3), 0.97, 1.02 ($2 \times$ d, 6H, $2 \times$ Me), 1.88 (hept, 2H, $J = 7.0$ Hz, CH_2), 3.05 (m, 2H, CH_2), 4.38, 4.82 ($2 \times$ d, 2H, $J = 10.0$ Hz, CH_2), 7.50–8.08 (m, 7H, H-arom.) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 14.37 (CH_3), 19.91, 19.98 ($2 \times CH_3$), 28.82 (CH), 62.89 (CH_2), 72.84 (CH_2), 118.23, 125.03, 125.26, 126.98, 127.08, 128.35, 129.50, 129.98, 130.62, 133.82 (C-arom.), 147.89 (C-6), 152.74 (C-2), 164.64 (C-4) ppm.

General procedure for the preparation of 1-(methylthiomethyl)-5-alkyl-6-(naphth-1-yl)-uracils **6d**,**f**

The uracils **5a**,**b** (0.84 g, 3 mmol) were silylated by refluxing in $10\,cm^3$ 1,1,1,3,3,3-*HMDS* in the presence of 15 mg $(NH_4)_2SO_4$. After evaporation of the solvent, the resulting solid was dissolved in anhydrous MeCN ($10\,cm^3$). The mixture was cooled to −40°C, and *TMS*-triflate (0.667 g, 3 mmol) was added in one portion, followed by dropwise addition of methylthiomethyl acetate (420 mg, 3.5 mmol). The temperature of the reaction mixture was raised gradually to −5°C, and stirring was continued at this temperature overnight. The reaction was quenched by adding cold saturated aqueous $NaHCO_3$ ($15\,cm^3$). The mixture was evaporated *in vacuo* to near dryness. EtOH ($2 \times 50\,cm^3$) was added, and the resulting suspension was evaporated *in vacuo* one more time. The solid was triturated with $CHCl_3$, and the solvent was removed *in vacuo* to afford a slightly yellow foam. Purification by silica gel column chromatography ($CHCl_3$) gave the acyclic nucleosides **6d**,**f**.

5-Isopropyl-1-(methylthiomethyl)-6-(naphth-1-yl)-uracil (**6d**; $C_{19}H_{20}N_2O_2S$)

Yield: 0.57 g (71%); m.p.: 185°C; MS (EI): $m/z = 340$ (M^+); ^{1}H NMR (*DMSO*-d_6, δ, 250 MHz): 0.96, 1.04 ($2 \times$ d, 6H, $J = 7.0$, $2 \times CH_3$), 1.90 (m, s, 4H, CH_3, CH), 3.90, 4.70 ($2 \times$ d, 2H, $J = 14.1$ Hz, CH_2), 7.53–8.08 (m, 7H, H-arom.) ppm; ^{13}C NMR (*DMSO*-d_6, δ, 75 MHz): 15.82 (CH_3), 19.17, 19.96 ($2 \times CH_3$), 29.07 (CH), 48.83 (CH_2), 118.41 (C-5), 124.57, 125.44, 126.96, 127.45, 127.59, 128.01, 129.49, 130.01, 132.98, 133.02 (C-arom.), 147.76 (C-6), 151.74 (C-2), 163.40 (C-4) ppm.

5-Ethyl-1-(methylthiomethyl)-6-(naphth-1-yl)-uracil (**6f**; $C_{18}H_{18}N_2O_2S$)

Yield: 0.6 g (77%); m.p.: 170°C; MS (EI): $m/z = 326$ (M^+); ^1H NMR (*DMSO*-d$_6$, δ, 250 MHz): 0.84 (t, 3H, $J = 7.4$ Hz, CH_3), 1.85, 2.12 ($2 \times$ m, 2H, CH_2), 2.16 (s, 3H, CH_3), 3.98, 4.96 ($2 \times$ d, 2H, $J = 14.2$ Hz, CH_2), 7.22–7.98 (m, 7H, H-arom.) ppm; ^{13}C NMR (*DMSO*-d$_6$, δ, 75 MHz): 13.54 (CH_3), 16.72 (CH_3), 20.11 (CH_2), 49.44 (CH_2), 117.70 (C-5), 124.19, 125.18, 127.06, 127.83, 128.02, 128.50, 128.88, 130.25, 130.57, 133.52 (C-arom.), 149.49 (C-6), 151.64 (C-2), 163.34 (C-4) ppm.

5-Ethyl-1-((hydroxyethoxy)-methyl)-6-(naphth-1-yl)-uracil (**6e**; $C_{19}H_{20}N_2O_4$)

Method A: Uracil **5a** (0.84 g, 3 mmol) was silylated by being refluxing in 10 cm^3 *HMDS* in the presence of 15 mg $(NH_4)_2SO_4$. After evaporation of the solvent, the resulting solid was dissolved in anhydrous MeCN (10 cm^3). The mixture was cooled to −40°C, and *TMS*-triflate (0.667 g, 3 mmol) was added in one portion, followed by dropwise addition of 1,3-dioxolane (3 mmol). The temperature of the mixture was raised gradually to −5°C, and stirring was continued at this temperature overnight. The reaction was quenched by addition cold saturated aqueous $NaHCO_3$ (15 cm^3). The mixture was evaporated *in vacuo* to near dryness. EtOH (2×50 cm^3) was added, and the resulting suspension was evaporated *in vacuo* one more time. The solid was triturated with $CHCl_3$, and the solvent was removed *in vacuo* to afford a slightly yellow foam. Purification by silica gel column chromatography ($CHCl_3$) gave the acyclic nucleoside **6e**.

Method B: To a solution of **7** (0.40 g, 0.016 mol) in MeOH (10 cm^3), 2 cm^3 of 1 *N* NaOMe in MeOH were added. After 2 h at room temperature, the *pH* was adjusted to 4.0 with 1 *N* HCl, and the solvent was evaporated under reduced pressure to obtain a solid which was purified by silica gel column chromatography (20% MeOH in $CHCl_3$) to give the acyclic nucleoside **6e**.

Yield: 0.75 g (73%); m.p.: 145°C; MS (EI): $m/z = 340$ (M^+); ^1H NMR ($CDCl_3$, δ, 250 MHz): 0.74 (t, 3H, $J = 7.4$ Hz, CH_3), 1.74, 2.01 ($2 \times$ m, 2H, CH_2), 3.16, 3.27 ($2 \times$ m, 2H, CH_2), 3.43 (m, 2H, CH_2), 4.50, 4.96 ($2 \times$ d, 2H, $J = 9.8$ Hz, CH_2), 5.27 (br, 1H, OH), 7.39–7.93 (m, 7H, H-arom.) ppm; ^{13}C NMR ($CDCl_3$, δ, 75 MHz): 13.44 (CH_3), 20.03 (CH_2), 61.11 (CH_2), 70.11 (CH_2), 73.50 (CH_2), 117.53 (C-5), 124.76, 125.15, 126.87, 127.40, 127.48, 128.67, 128.97, 130.27, 130.73, 133.40 (C-arom.), 149.19 (C-6), 153.46 (C-2), 165.22 (C-4) ppm.

1-((2-Acetoxyethoxy)-methyl)-5-ethyl-6-(naphth-1-yl)-uracil (**7**; $C_{21}H_{22}N_2O_5$)

N,O-*Bis*-(trimethylsilyl)-acetamide (6 cm^3, 0.024 mol) was added dropwise under nitrogen to a stirred mixture of **5a** (2.66 g, 0.01 mol) and acetoxyethyl acetoxymethyl ether (2.7 g, 0.015 mol) in CH_2Cl_2 (25 cm^3). After 3 h of stirring at room temperature, the clear solution was cooled to 0°C, and $SnCl_4$ (0.2 cm^3, 0.002 mol) was added. The mixture was then warmed to room temperature, left stirring overnight, and finally poured slowly into a mixture of cold saturated aqueous $NaHCO_3$ (50 cm^3) and $CHCl_3$ (100 cm^3). The resulting emulsion was separated by filtration through Celite, the aqueous layer was extracted further with EtOAc (3×50 cm^3), and the combined organic layers were dried over anhydrous Na_2SO_4 and evaporated under reduced pressure. Trituration of the remaining oily residue with Et_2O afforded the product as colorless crystals.

Yield: 1.9 g (66%); m.p.: 90°C; MS (EI): $m/z = 382$ (M^+); ^1H NMR ($CDCl_3$, δ, 250 MHz): 0.76 (t, 3H, $J = 7.1$ Hz, CH_3), 1.75, 2.01 ($2 \times$ m, 2H, CH_2), 1.91 (s, 3H, CH_3), 3.43 (m, 2H, CH_2), 3.89 (m, 2H, CH_2), 4.42, 5.09 ($2 \times$ d, 2H, $J = 10.1$ Hz, CH_2), 7.39–7.93 (m, 7H, H-arom.) ppm; ^{13}C NMR ($CDCl_3$, δ, 75 MHz): 13.42 (CH_3), 19.93 (CH_3), 20.69 (CH_2), 63.20 (CH_2), 66.93 (CH_2), 73.92 (CH_2), 117.64 (C-5), 124.47, 124.64, 125.04, 126.82, 127.47, 127.68, 128.70, 130.32, 130.60, 133.38 (C-arom.), 149.56 (C-6), 152.33 (C-2), 163.95 (C-4), 170.87 (C=O) ppm.

Virus and cells

The HIV-1 strain HTLV-IIIB [12] and the NNRTI resistant strain N119 [13] were propagated in H9 cells [14] at 37°C, 5% CO_2 using RPMI 1640 with 10% heat-inactivated fetal calf serum (FCS) and antibiotics (growth medium). The culture supernatant was filtered (0.45 nm), aliquoted, and stored at −80°C until use. Both HIV-1 strains were obtained from the NIH AIDS Research and Reference Program.

Inhibition of HIV-1 replication

Compounds were examined for possible antiviral activity against both strains of HIV-1 using MT4 cells as target cells. MT4 cells were incubated with virus (0.005 MOI) for 2 h, washed, and thereafter added in a proportion of 1:10 to uninfected cells which had been preincubated in growth medium containing the test compound for 6 days in parallel with virus-infected control cultures without compound added. Expression of HIV in the culture medium was quantitated by HIV-1 antigen detection assay ELISA [15]. Compounds mediating less than 30% reduction of antigen expression were considered without biological activity. Compounds mediating a reduction of 30% or more were examined for cytotoxic effect using the concentration dependent inhibition of MT-4 cell proliferation as a measure of cytotoxicity employing the MTT assay as previously described [16]. A 30% inhibition of cell growth relative to control cultures was considered significant.

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Received September 25, 2001. Accepted (revised) December 3, 2001

Verleger: Springer-Verlag KG, Sachsenplatz 4–6, A-1201 Wien. – Herausgeber: Österreichische Akademie der Wissenschaften, Dr.-Ignaz-Seipel-Platz 2, A-1010 Wien, und Gesellschaft Österreichischer Chemiker, Eschenbachgasse 9, A-1010 Wien. – Redaktion: Währinger Straße 38, A-1090 Wien. – Satz und Umbruch: Thomson Press Ltd., New Delhi, India. – Offsetdruck: Manz Crossmedia, A-1051 Wien. – Verlagsort: Wien. – Herstellungsort: Wien. – Printed in Austria.