Synthesis of S^2-Alkyl-2-thiouridines

Adel A.-H. Abdel-Rahman,[a] Magdy A. Zahran,[a] Ahmed E.-S. Abdel-Megied,[a] Erik B. Pedersen,*[a] Claus Nielsen[b]

[a] Department of Chemistry, Odense University, Campusvej 55, DK-5230 Odense M, Denmark

[b] Retrovirus Laboratory, Department of Virology, Statens Seruminstitut, Artillerivej 5, DK-2300 Copenhagen, Denmark

Fax +4566158780; E-mail EBP@OUKEMI.OU.DK

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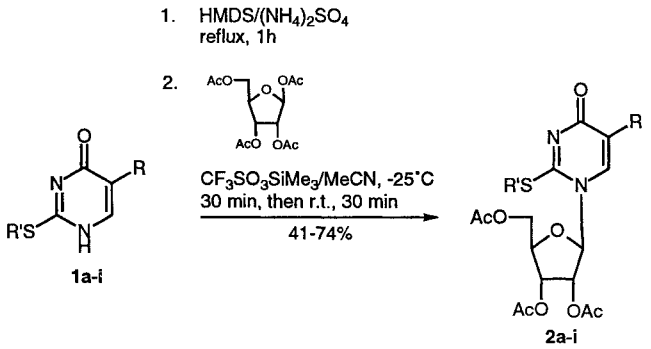
5-Substituted S^2-alkyl-2-thiouracils **1a–i** were treated with 1,1,1,3,3,3-hexamethyldisilazane and ammonium sulfate at reflux temperature and condensed with 1,2,3,5-tetra-*O*-acetyl-β-D-ribofuranose in acetonitrile using trimethylsilyl trifluoromethanesulfonate as a catalyst to afford the corresponding protected nucleosides **2a–i** which were deprotected with saturated ammonia in methanol. When the nucleobase was substituted with ethoxycarbonyl or cyano groups in the 5-position or was unsubstituted, the deprotection reaction of the nucleoside also resulted in replacement of the methylthio group. This was not observed with 5-alkyl and 5-methoxy substituents.

Singer et al.[1] reported that O^2-ethyl and O^4-methyl derivatives of uridine 5′-diphosphate (UDP) could be copolymerized with UDP or cytidine 5′-diphosphate (CDP), using polynucleotide phosphorylase. These copolymers were used as templates for DNA dependent RNA polymerase in the presence of Mn^{2+}. O^2-Alkylation of uracil (U) or thymine (T) is, in contrast to O^4-alkylation, a relatively frequent result of treatment of double-stranded nucleic acids with *N*-nitroso alkylating agents. In single-stranded nucleic acids both O^2- and O^4-alkylation of U may be involved in the high carcinogenicity of these alkylating agents.[2–6]

The reaction of nucleic acids with carcinogens of *N*-nitroso type showed that O^2-alkylpyrimidines were the major products.[5] Only a few examples of the corresponding S^2-alkylated 2-thiouridines have been reported[7–9] and they were typically prepared by alkylation of the corresponding 2-thio nucleoside. We thought that such compounds could be synthesized in a more straightforward manner by condensing S^2-alkylated 2-thiouracils with appropriate sugar derivatives. Considering the interest in O^2-methyl nucleosides in conjunction with DNA as mentioned above, we focused this work on the synthesis of S^2-alkylated 2-thiouridine derivatives.

In the present investigation we have synthesized S^2-methyl as well as the S^2-ethyl-2-thiouracils **1** by alkylation of the appropriate 2-thiouracils according to the procedure of Brown et al.[10] and treated them according to the procedure of Wittenburg[11] with 1,1,1,3,3,3-hexamethyldisilazane and ammonium sulfate at reflux temperature in order to obtain the silylated derivatives which where condensed with 1,2,3,5-tetra-*O*-acetyl-β-D-ribofuranose in dry acetonitrile using trimethylsilyl trifluoromethanesulfonate as the catalyst according to the method of Vorbrüggen et al.[12] After chromatographic purification the β-anomers **2a–i** were obtained in 61–93% yield. Deprotection of the hydroxy groups in the nucleosides **2a–g** with ammonia in methanol afforded the corresponding S^2-alkyl-2-thiouridine derivatives **3a–g** in 40–96% yield after chromatographic purification.

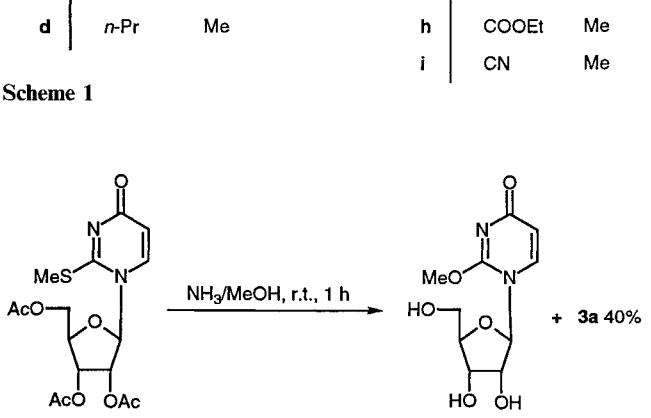
The methylthio group was very susceptible to a nucleophilic substitution reaction and was in some cases replaced by a methoxy group. On deprotection of **2a** we obtained **3a** in 40% yield together with **4** in 34% yield. When deprotection of **2a** was performed with sodium methoxide in methanol, we only isolated the O^2-methylated compound **4** in 43% yield. Substitution of the meth-



1-3	R	R'
a	H	Me
b	Me	Me
c	Et	Me
d	*n*-Pr	Me

1-3	R	R'
e	CH_2Ph	Me
f	Me	Et
g	OMe	Et
h	COOEt	Me
i	CN	Me

Scheme 1



Scheme 2

Table 1. Yields and Physical Data of the New Compounds Prepared

Compound	Yield (%)	mp (°C)	$[\alpha]_D^{34}$[a]	Molecular Formula[b]	MS *m/z* (%)
2a	93				
2b	90				
2c	91				
2d	85				
2e	76				
2f	93				429 ($M + H^+$)[c]
2g	91				445 ($M + H^+$)[c]
2h	61				
2i	70				
3a	40	152–153		$C_{10}H_{14}N_2O_5S$ (274.3)	274 (M^+, 0.4)
3b	86	204–205	+25.73	$C_{11}H_{16}N_2O_5S$ (288.3)	288 (M^+, 0.7)
3c	95	202–203	+33.33	$C_{12}H_{18}N_2O_5S \cdot 0.5H_2O$ (311.4)	302 (M^+, 2)
3d	85	183–184		$C_{13}H_{20}N_2O_5S$ (316.3)	316 (M^+, 3)
3e	80	132–134	+35.59	$C_{17}H_{20}N_2O_5S$ (364.1093)	364 (M^+, 0.7)
3f	73	232–233	+24.33	$C_{12}H_{18}N_2O_5S$ (302.4)	302 (M^+, 0.7)
3g	77			$C_{12}H_{18}N_2O_6S$ (318.4)	318 (M^+, 0.05)

[a] Specific rotation **(3b, c, 3e, f)** was measured in MeOH; $c = 1$.
[b] The microanalyses **(3a–3d, 3f–3g)** or HRMS **(3e)** data were in satisfactory agreement with the calculated values: C ± 0.44, H ± 0.32, N ± 0.45; $m/z + 0.0004$ (M^+).
[c] FAB MS **(2f, g)** was measured in DMSO + 3-nitrobenzyl alcohol.

Table 2. NMR Data of the New Compounds Prepared

Compound	^{1}H NMR (DMSO-d_6/TMS) δ, J (Hz)	^{13}C NMR (DMSO-d_6/TMS) δ
2a	2.07 (s, 6H, 2 × $COCH_3$), 2.11 (s, 3H, $COCH_3$), 2.51 (s, 3H, SCH_3), 4.34 (d, 2H, J = 4.1, 5′-H), 4.41 (m, 1H, 4′-H), 5.31 (t, 1H, J = 6.1, 3′-H), 5.43 (t, 1H, J = 6.1, 2′-H), 5.97 (d, 1H, J = 5.7, 1′-H), 6.05 (d, 1H, J = 7.8, 5-H), 7.88 (d, 1H, J = 7.7, 6-H)	14.20 (SCH_3), 19.95, 20.15, 20.37 (3 × $COCH_3$), 62.68 (C-5′), 69.26 (C-3′), 72.07 (C-2′), 79.76 (C-4′), 88.92 (C-1′), 109.31 (C-5), 138.95 (C-6), 161.86 (C-2), 165.92 (C-4), 168.94, 169.20, 169.82 (3 × $COCH_3$)
2b	1.86 (s, 3H, 5-CH_3), 2.07 (s, 3H, $COCH_3$), 2.08 (s, 3H, $COCH_3$), 2.11 (s, 3H, $COCH_3$), 2.51 (s, 3H, SCH_3), 4.38 (m, 3H, 4′-H, 5′-H), 5.33 (m, 1H, 3′-H), 5.43 (t, 1H, J = 6.2, 2′-H), 5.96 (d, 1H, J = 5.9, 1′-H), 7.78 (s, 1H, 6-H)	13.22 (SCH_3), 14.26 (5-CH_3), 19.97, 20.16, 20.39 (3 × $COCH_3$), 62.70 (C-5′), 69.21 (C-3′), 72.04 (C-2′), 79.71 (C-4′), 88.90 (C-1′), 117.85 (C-5), 134.81 (C-6), 160.80 (C-2), 167.00 (C-4), 168.94, 169.21, 169.82 (3 × $COCH_3$)
2c	1.17 (t, 3H, J = 7.3, CH_2CH_3), 2.11 (s, 3H, $COCH_3$), 2.15 (s, 3H, $COCH_3$), 2.18 (s, 3H, $COCH_3$), 2.44 (q, 2H, J = 7.3, CH_2CH_3), 2.62 (s, 3H, SCH_3), 4.33 (d, 1H, J = 12.2, 5′-H), 4.40 (br s, 1H, 4′-H), 4.44 (dd, 1H, J = 12.5, 2.9, 5′-H), 5.31 (m, 2H, 2′-H, 3′-H), 6.06 (d, 1H, J = 4.9, 1′-H), 7.37 (s, 1H, 6-H)	12.04 (CH_2CH_3), 14.77 (SCH_3), 20.12, 20.21, 20.31 (3 × $COCH_3$), 21.20 (CH_2CH_3), 62.82 (C-5′), 69.84 (C-3′), 72.82 (C-2′), 80.44 (C-4′), 88.35 (C-1′), 124.74 (C-5), 131.82 (C-6), 161.21, (C-2), 167.87 (C-4), 168.90, 169.32, 169.72 (3 × $COCH_3$)
2d	0.87 (t, 3H, J = 7.2, CH_3), 1.48 (hextet, 2H, J = 7.3, CH_2), 2.06 (s, 3H, $COCH_3$), 2.08 (s, 3H, $COCH_3$), 2.11 (s, 3H, $COCH_3$), 2.25 (t, 2H, J = 7.8, CH_2), 2.51 (s, 3H, SCH_3), 4.38 (m, 3H, 4′-H, 5′-H), 5.35 (m, 1H, 3′-H), 5.46 (t, 1H, J = 6.2, 2′-H), 5.96 (d, 1H, J = 6.2, 1′-H), 7.67 (s, 1H, 6-H)	13.34 (CH_3), 14.24 (SCH_3), 19.94, 20.15, 20.35 (3 × $COCH_3$), 20.80 (CH_2), 29.28 (CH_2), 62.72 (C-5′), 69.31 (C-3′), 71.90 (C-2′), 79.80 (C-4′), 88.85 (C-1′), 121.54 (C-5), 134.69 (C-6), 160.66 (C-2), 166.45 (C-4), 168.91, 169.21, 169.76 (3 × $COCH_3$)
2e	1.96 (s, 3H, $COCH_3$), 2.08 (s, 3H, $COCH_3$), 2.12 (s, 3H, $COCH_3$), 2.61 (s, 3H, SCH_3), 3.74 (d, 1H, J = 16.3, CH_2), 3.77 (d, 1H, J = 16.3, CH_2), 4.03 (dd, 1H, J = 12.4, 2.6, 5′-H), 4.22 (dd, 1H, J = 12.4, 4.0, 5′-H), 4.31 (m, 1H, 4′-H), 5.15 (m, 2H, 2′-H, 3′-H), 6.01 (d, 1H, J = 5.3, 1′-H), 7.21–7.32 (m, 6H, H_{arom}, 6-H)	14.71 (SCH_3), 20.01, 20.19, 20.34 (3 × $COCH_3$), 33.48 (CH_2), 62.73 (C-5′), 69.97 (C-3′), 72.74 (C-2′), 80.30 (C-4′), 88.47 (C-1′), 123.00 (C-5), 126.46, 128.34, 128.88 (C_{arom}), 133.68 (C-6), 137.71 (C_{arom}), 161.33 (C-2), 167.51 (C-4), 168.71, 169.10, 169.54 (3 × $COCH_3$)
2f	1.28 (t, 3H, J = 7.5, SCH_2CH_3), 1.86 (s, 3H, CH_3), 2.06 (s, 3H, $COCH_3$), 2.08 (s, 3H, $COCH_3$), 2.11 (s, 3H, $COCH_3$), 3.14 (q, 2H, J = 7.5, SCH_2), 4.36 (m, 3H, 4′-H, 5′-H), 5.32 (d, 1H, J = 2.5, 3′-H), 5.42 (m, 1H, 2′-H), 5.93 (d, 1H, J = 5.0, 1′-H), 7.77 (s, 1H, 6-H)	13.25 (5-CH_3), 13.96 (SCH_2CH_3), 19.96, 20.16, 20.39 (3 × $COCH_3$), 25.74 (SCH_2), 62.71 (C-5′), 69.26 (C-3′), 71.99 (C-2′), 79.74 (C-4′), 88.75 (C-1′), 117.98 (C-5), 134.86 (C-6), 160.07 (C-2), 167.02 (C-4), 168.92, 169.23, 169.81 (3 × $COCH_3$)

Table. (continued)

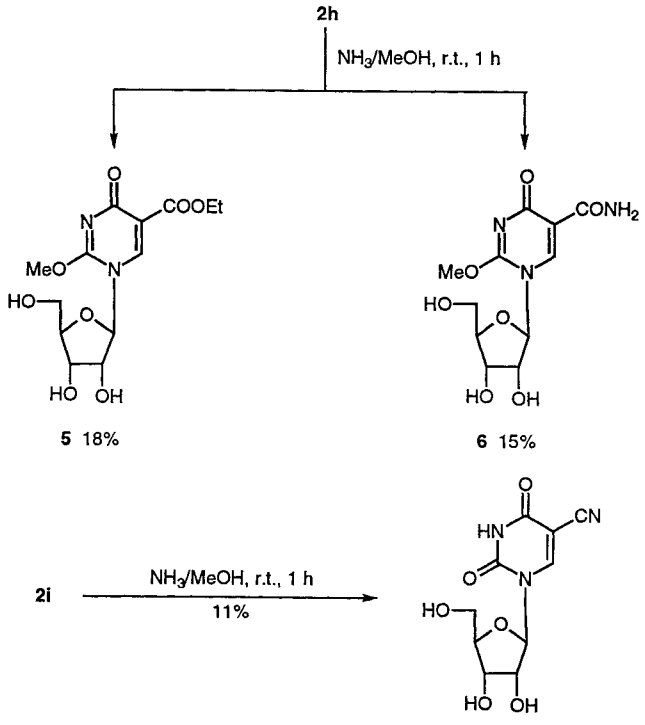
Compound	^{1}H NMR (DMSO-d_6/TMS) δ, J (Hz)	^{13}C NMR (DMSO-d_6/TMS) δ
2g	1.27 (t, 3H, J = 7.5, SCH_2CH_3), 2.05, 2.07, 2.11 (3 × s, 9H, 3 × $COCH_3$), 3.12 (q, 2H, J = 7.5, SCH_2), 3.70 (s, 3H, OCH_3), 4.42 (s, 3H, 4′-H, 5′-H), 5.32 (m, 1H, 3′-H), 5.51 (t, 1H, J = 7.5, 2′-H), 6.00 (d, 1H, J = 7.5, 1′-H), 7.35 (s, 1H, 6-H)	14.04 (SCH_2CH_3), 19.97, 20.20, 20.39 (3 × $COCH_3$), 26.00 (SCH_2), 55.95 (OCH_3), 62.74 (C-5′), 69.24 (C-3′), 71.75 (C-2′), 80.07 (C-4′), 88.94 (C-1′), 117.81 (C-5), 142.06 (C-6), 157.01 (C-2), 161.69 (C-4), 168.90, 169.27, 169.87 (3 × $COCH_3$)
2h	1.23 (t, 3H, J = 6.9, CH_3), 2.05 (s, 3H, $COCH_3$), 2.08 (s, 3H, $COCH_3$), 2.09 (s, 3H, $COCH_3$), 2.52 (s, 3H, SCH_3), 4.21 (m, 2H, 5′-H), 4.35 (m, 2H, CH_2), 4.46 (m, 1H, 4′-H), 5.34 (t, 1H, J = 6.1, 3′-H), 5.46 (t, 1H, J = 5.8, 2′-H), 6.00 (d, 1H, J = 5.6, 1′-H), 8.40 (s, 1H, 6-H)	13.96 (CH_3), 14.62 (SCH_3), 20.07, 20.24, 20.33 (3 × $COCH_3$), 60.63 (OCH_2), 62.61 (C-5′), 69.37 (C-3′), 72.87 (C-2′), 80.40 (C-4′), 89.26 (C-1′), 111.49 (C-5), 143.48 (C-6), 161.60 (C-2), 161.80 (C-4), 163.60 ($COOEt$), 169.03, 169.23, 169.91 (3 × $COCH_3$)
2i	2.07 (s, 3H, $COCH_3$), 2.08 (s, 3H, $COCH_3$), 2.10 (s, 3H, $COCH_3$), 2.54 (s, 3H, SCH_3), 4.39 (m, 3H, 4′-H, 5′-H), 5.35 (t, 1H, J = 6.0, 3′-H), 5.51 (t, 1H, J = 4.5, 2′-H), 6.02 (d, 1H, J = 4.2, 1′-H), 8.72 (s, 1H, 6-H)	14.75 (SCH_3), 20.04, 20.15, 20.44 (3 × $COCH_3$), 62.30 (C-5′), 68.30 (C-3′), 72.89 (C-2′), 79.79 (C-4′), 90.00 (C-1′), 95.57 (CN), 114.54 (C-5), 147.87 (C-6), 161.99 (C-2), 163.89 (C-4), 168.93, 169.04, 169.85 (3 × $COCH_3$)
3a	2.48 (s, 3H, SCH_3), 3.61 (m, 2H, 5′-H), 3.94 (m, 1H, 4′-H), 4.01 (m, 1H, 3′-H), 4.11 (q, 1H, J = 5.2, 2′-H), 5.23 (m, 2H, 3′-OH, 5′-OH), 5.58 (d, 1H, J = 5.9, 2′-OH), 5.70 (d, 1H, J = 5.3, 1′-H), 5.95 (d, 1H, J = 7.7, 5-H), 8.08 (d, 1H, J = 7.8, 6-H)	14.07 (SCH_3), 60.60 (C-5′), 69.86 (C-3′), 74.52 (C-2′), 85.81 (C-4′), 91.31 (C-1′), 108.73 (C-5), 139.16 (C-6), 162.28 (C-2), 166.53 (C-4)
3b	1.84 (s, 3H, CH_3), 2.50 (s, 3H, SCH_3), 3.66 (m, 2H, 5′-H), 3.96 (m, 1H, 4′-H), 4.06 (m, 1H, 3′-H), 4.14 (m, 1H, 2′-H), 5.30 (m, 2H, 3′-OH, 5′-OH), 5.62 (d, 1H, J = 5.7, 2′-OH), 5.70 (d, 1H, J = 5.3, 1′-H), 8.02 (s, 1H, 6-H)	13.46 (CH_3), 14.21 (SCH_3), 60.71 (C-5′), 69.91 (C-3′), 73.43 (C-2′), 85.82 (C-4′), 91.31 (C-1′), 117.24 (C-5), 135.39 (C-6), 161.21 (C-2), 167.74 (C-4)
3c	1.05 (t, 3H, J = 7.4, CH_3), 2.26 (q, 2H, J = 7.4, CH_2), 2.49 (s, 3H, SCH_3), 3.65 (m, 2H, 5′-H), 3.96 (m, 1H, 4′-H), 4.04 (q, 1H, J = 4.1, 3′-H), 4.14 (q, 1H, J = 5.3, 2′-H), 5.26 (m, 2H, 3′-OH, 5′-OH), 5.60 (d, 1H, J = 5.8, 2′-OH), 5.70 (d, 1H, J = 5.3, 1′-H), 8.01 (s, 1H, 6-H)	12.11 (CH_3), 14.19 (SCH_3), 20.59 (CH_2), 60.59 (C-5′), 69.96 (C-3′), 74.66 (C-2′), 85.79 (C-4′), 91.47 (C-1′), 122.66 (C-5), 134.53 (C-6), 160.87 (C-2), 167.13 (C-4)
3d	0.86 (t, 3H, J = 7.2, CH_3), 1.48 (hextet, 2H, J = 7.2, CH_2), 2.22 (m, 2H, CH_2), 2.48 (s, 3H, SCH_3), 3.68 (m, 2H, 5′-H), 3.95 (m, 1H, 4′-H), 4.03 (m, 1H, 3′-H), 4.13 (q, 1H, J = 5.1, 2′-H), 5.21 (m, 2H, 3′-OH, 5′-OH), 5.56 (d, 1H, J = 5.7, 2′-OH), 5.69 (d, 1H, J = 5.3, 1′-H), 7.99 (s, 1H, 6-H)	13.49 (CH_3), 14.08 (SCH_3), 20.30 (CH_2), 29.26 (CH_2), 60.57 (C-5′), 69.89 (C-3′), 74.61 (C-2′), 85.73 (C-4′), 91.37 (C-1′), 120.86 (C-5), 135.08 (C-6), 160.74 (C-2), 167.09 (C-4)
3e	2.47 (s, 3H, SCH_3), 3.58 (m, 4H, CH_2, 5′-H), 3.94 (m, 1H, 4′-H), 4.00 (m, 1H, 3′-H), 4.10 (m, 1H, 2′-H), 5.21 (br s, 2H, 3′-OH, 5′-OH), 5.61 (br s, 1H, 2′-OH), 5.68 (d, 1H, J = 5.0, 1′-H), 7.23 (m, 5H, H_{arom}), 8.09 (s, 1H, 6-H)	14.14 (SCH_3), 33.14 (CH_2), 60.62 (C-5′), 69.83 (C-3′), 74.57 (C-2′), 85.72 (C-4′), 91.44 (C-1′), 120.54 (C-5), 125.89, 128.08, 128.35, 139.43 (C_{arom}), 136.16 (C-6), 161.19 (C-2), 166.71 (C-4)
3f	1.28 (t, 3H, J = 7.5, SCH_2CH_3), 1.82 (s, 3H, 5-CH_3), 3.13 (q, 2H, J = 7.5, SCH_2), 3.63 (m, 2H, 5′-H), 3.92 (q, 1H, J = 3.1, 4′-H), 4.02 (q, 1H, J = 3.7, 3′-H), 4.12 (q, 1H, J = 5.3, 2′-H), 5.20 (m, 2H, 3′-OH, 5′-OH), 5.53 (d, 1H, J = 5.8, 2′-OH), 5.67 (d, 1H, J = 5.5, 1′-H), 7.99 (s, 1H, 6-H)	13.43 (5-CH_3), 13.97 (CH_3), 25.50 (SCH_2), 60.67 (C-5′), 69.85 (C-3′), 74.28 (C-2′), 85.73 (C-4′), 91.11 (C-1′), 117.28 (C-5), 135.37 (C-6), 160.48 (C-2), 167.59 (C-4)
3g	1.28 (t, 3H, J = 7.3, SCH_2CH_3), 3.13 (q, 2H, J = 7.3, SCH_2), 3.61 (s, 3H, OCH_3), 3.67 (m, 2H, 5′-H), 3.97 (m, 1H, 4′-H), 4.06 (m, 1H, 3′-H), 4.16 (m, 1H, 2′-H), 5.21 (m, 1H, 3′-OH), 5.37 (m, 1H, 5′-OH), 5.56 (m, 1H, 2′-OH), 5.73 (d, 1H, J = 4.8, 1′-H), 7.94 (s, 1H, 6-H)	14.06 (SCH_2CH_3), 25.65 (SCH_2), 55.66 (OCH_3), 60.22 (C-5′), 69.73 (C-3′), 74.84 (C-2′), 85.62 (C-4′), 91.91 (C-1′), 118.17 (C-5) 142.04 (C-6), 156.54 (C-2), 162.08 (C-4)

ylthio group was also a concomitant reaction during deprotection of **2h**. When **2i** was deprotected, it was not possible to detect the corresponding O^2-methyl derivative. The substitution reaction was followed by a demethylation reaction of the methoxy group affording 5-cyanouridine **7** in 11% yield. Also hydrolysis by water present in the methanol could explain the formation of compound **7**.

Replacement of the methylthio group during the deprotection reaction was not observed when 5-alkyl- or 5-methoxy substituents were present in the uracil ring, **2b–g**, because of their electron-donating effect which retards the aromatic nucleophilic substitution reaction.

The compounds **3a–e, 4–7** were tested for their antiviral activity against herpes simplex virus (HSV-1) in Vero cells and against human immunodeficiency virus (HIV-1) in MT-4 cells, however, no activity was observed.

Anhyd MeCN was distilled from P_2O_5 followed by distillation from CaH_2. All other solvents were used after distillation. Analytical silica gel TLC plates 60 F_{254} and silica gel (230–400 mesh) were purchased from Merck. Optical rotations were measured with a Perkin-Elmer 141 polarimeter. NMR spectra were recorded on a Bruker AC 250 FT NMR spectrometer at 250 MHz for ^{1}H NMR and 62.9 MHz for ^{13}C NMR with TMS as an internal standard. EI mass spectra were obtained on a Varian MAT 311 A spectrometer. FAB mass spectra were recorded on a Kratos MS-50 spectrometer.



Scheme 3

Preparation of 2-Ethylthiopyrimidin-4(1*H*)-ones 1f, g; General Procedure:

A solution of substituted 2-thiouracils (0.1 mol), EtI (0.1 mol) and 1 M aq NaOH (100 mL) and EtOH (200 mL) was stirred at 60 °C overnight. Half of the EtOH was evaporated off and the mixture allowed to cool. The solid was collected, dried and recrystallized from EtOH.

2-Ethylthio-5-methylpyrimidin-4(1H)-one (**1f**); yield: 72 %; colorless needles; mp 158 °C.

$C_7H_{10}N_2OS$ (170.2) calc. C 49.39 H 5.92 N 16.45
found 49.16 5.92 16.24

^{1}H NMR (DMSO-d_6): δ = 1.28 (t, 3 H, J = 7.5 Hz, SCH_2CH_3), 1.87 (s, 3 H, 5-CH_3), 3.07 (q, 2 H, J = 7.5 Hz, SCH_2CH_3), 7.72 (s, 1 H, 6-H), 12.56 (s, 1 H, NH).

^{13}C NMR (DMSO-d_6): δ = 12.29 (5-CH_3), 14.37 (CH_2CH_3), 23.95 (CH_2CH_3), 118.74 (C-5), 150.27 (C-6), 158.73 (C-4), 162.89 (C-2).

2-Ethylthio-5-methoxypyrimidine-4(1H)-one (**1g**); yield: 68 %; colorless needles; mp > 300 °C.

$C_7H_{10}N_2O_2S$ (176.2) calc. C 45.14 H 5.41 N 15.04
found 45.01 5.39 14.98

^{1}H NMR (DMSO-d_6): δ = 1.29 (t, 3 H, J = 7.5 Hz, SCH_2CH_3), 3.07 (q, 2 H, J = 7.5 Hz, SCH_2CH_3), 3.70 (s, 3 H, OCH_3), 7.52 (s, 1 H, 6-H).

^{13}C NMR (DMSO-d_6): δ = 14.42 (CH_2CH_3), 24.25 (CH_2CH_3), 56.10 (OCH_3), 130.54 (C-5), 143.55 (C-6), 151.23 (C-4), 157.87 (C-2).

Preparation of 2a–i; General Procedure:

2-Alkylthiopyrimidin-4(1*H*)-one derivatives **1a–i** (8 mmol) were treated with 1,1,1,3,3,3-hexamethyldisilazane (30 mL) and $(NH_4)_2SO_4$ (50 mg) at reflux for 1 h (clear solution after 0.5 h). The solvent was removed in vacuo. The residue was dissolved in anhyd MeCN (20 mL) and 1,2,3,5-tetra-*O*-acetyl-β-D-ribofuranose (1.71 g, 5.4 mmol) was added. The reaction mixture was cooled to − 30 °C. A solution of $CF_3SO_3SiMe_3$ (1.3 mL, 6.5 mmol) in anhyd MeCN (5 mL) was added dropwise with stirring. The mixture was stirred for 0.5 h at − 25 °C and then at r.t. for 0.5 h. The mixture was diluted with CH_2Cl_2 (200 mL) and extracted with ice-cold sat. aq $NaHCO_3$. The organic phase was separated, washed with cold H_2O (3 × 150 mL), and dried (Na_2SO_4). The solvent was evaporated in vacuo to obtain the crude products which were chromatographed on a silica gel column using 5–8 % MeOH/CH_2Cl_2 as eluent.

Preparation of 3–7; General Procedure:

Sat. NH_3 in MeOH (40 mL) was added with stirring to a solution of **2a–i** (2–5 mmol) in MeOH (20 mL) at 0 °C. The mixture was stirred at r.t. for 1 h and the solvent was evaporated in vacuo. The residue was chromatographed on a silica gel column with 5 % MeOH/CH_2Cl_2 to give the products **3–7**. Compound **2a** (1.0 g, 2 mmol) was also treated with MeONa (2.2 mmol) in MeOH (30 mL) at 0 °C and stirring at r.t. for 1 h. After neutralization with NH_4Cl (120 mg, 2.3 mmol), the solvent was removed in vacuo and the residue was chromatographed on a silica gel column with 7 % MeOH/CH_2Cl_2 to give **4** in 43 % yield.

O^2-Methyluridine (**4**); yield: 0.43 g (34 %); mp 172–173 °C (Lit.[13–15] mp 173 °C); $[\alpha]_D^{34}$ + 35.45 (c = 1, H_2O).

Ethyl O^2-Methyluridine-5-carboxylate (**5**); yield: 200 mg (19 %); foam.

HRMS: $C_{13}H_{18}N_2O_8$, calc. 330.1063; found 330.1069.

^{1}H NMR (DMSO-d_6): δ = 1.24 (t, 3 H, J = 7.0 Hz, CH_3), 3.16 (s, 3 H, OCH_3), 3.60 (dd, 1 H, J = 12.0, 2.2 Hz, 5′-H), 3.73 (dd, 1 H, J = 12.0, 2.2 Hz, 5′-H), 3.92 (m, 1 H, 4′-H), 3.99 (t, 1 H, J = 4.6 Hz, 3′-H), 4.07 (t, 1 H, J = 3.6 Hz, 2′-H), 4.16 (q, 2 H, J = 7.1 Hz, OCH_2), 5.15 (br s, 3 H, 2′-OH, 3′-OH, 5′-OH), 5.80 (d, 1 H, J = 3.2 Hz, 1′-H), 8.94 (s, 1 H, 6-H).

^{13}C NMR (DMSO-d_6): δ = 13.93 (CH_3), 55.78 (OCH_3), 59.80, 59.96 (OCH_2, C-5′), 68.91 (C-3′), 74.58 (C-2′), 84.91 (C-4′), 90.49 (C-1′), 110.38 (C-5), 144.03 (C-6), 162.19 (C-2), 163.26 (C-4), 164.92 (*C*OEt).

MS EI: m/z (%) = 330 (M^+, 4).

O^2-Methyluridine-5-carboxamide (**6**); yield: 150 mg (15 %); mp 174–176 °C.

HRMS: $C_{11}H_{15}N_3O_7$ calc. C 301.091; found 301.090.

^{1}H NMR (DMSO-d_6): δ = 3.62 (m, 2 H, 5′-H), 3.96 (m, 5 H, OCH_3, 3′-H, 4′-H), 4.13 (br s, 1 H, 2′-H), 5.11 (m, 2 H, 3′-OH, 5′-OH), 5.53 (br s, 1 H, 2′-OH), 5.76 (d, 1 H, J = 4.5 Hz, 1′-H), 7.50 (s, 1 H, 6-H), 8.82 (s, 2 H, NH_2).

^{13}C NMR (DMSO-d_6): δ = 56.11 (OCH_3), 60.52 (C-5′), 69.52 (C-3′), 74.38 (C-2′), 85.35 (C-4′), 90.40 (C-1′), 111.30 (C-5), 143.74 (C-6), 155.42 (C-2), 163.69 (C-4), 168.95 (CO).

MS EI: m/z (%) = 301 (M^+, 0.7).

Uridine-5-carbonitrile (**7**); yield: 151 mg (11 %); mp 188–190 °C (Lit.[16–18], mp 191–192 °C).

(1) Singer, B.; Fraenkel-Conrat, H.; Kusmierek, J.T. *Proc. Nat. Acad. Sci. U.S.A.* **1978**, *75*, 1722.
(2) Singer, B.; Fraenkel-Conrat, H. *Biochemistry* **1975**, *14*, 772.
(3) Sun, L.; Singer, B. *Biochemistry* **1975**, *14*, 1795.
(4) Singer, B.; Kusmierek, J.T. *Biochemistry* **1976**, *15*, 5052.
(5) Singer, B. *Nature (London)* **1976**, *264*, 333.
(6) Singer, B. *J. Toxicol. Environ. Health* **1977**, *2*, 1279.
(7) Ogihara, T.; Mitsunobu, O. *Chem. Lett.* **1982**, 1621.
(8) Sato, E.; Machida, M.; Kanaoka, Y. *Chem. Pharm. Bull.* **1980**, *28*, 1722.
(9) Vorbrüggen, H.; Krolikiewicz, K. *Angew. Chem.* **1976**, *88*, 724.
(10) Brown, T.H.; Blakemore, R.C.; Durant, G.J.; Emmett, J.C.; Ganellin, C.R.; Parsons, M.E.; Rawlings, D.A.; Walker, T.F. *Eur. J. Med. Chem.* **1988**, *23*, 53.
(11) Wittenburg, E. *Z. Chem.* **1964**, *4*, 303.
(12) Vorbrüggen, H.; Krolikiewicz, K.; Bennua, B. *Chem. Ber.* **1981**, *114*, 1234.

(13) Brown, D.M.; Todd, Sir A.R.; Varadarajan, S. *J. Chem. Soc.* **1957**, 868.
(14) Kimura, J.; Fujisawa, Y.; Yoshizawa, T.; Fuknda, K.; Mitsunobu, O. *Bull. Chem. Soc. Jpn.* **1979**, *52*, 1191.
(15) Kimura, J.; Yagi, K.; Suzuki, H.; Mitsunobu, O. *Bull. Chem. Soc. Jpn.* **1980**, *53*, 3670.
(16) Prystas, M.; Sorm, F. *Coll. Czech. Chem. Commun.* **1966**, *31*, 3990.
(17) Torrence, P.F.; Bhooshan, B.; Descamps, J.; De Clercq, E. *J. Med. Chem.* **1977**, *20*, 974.
(18) Inoue, H.; Ueda, T. *Chem. Pharm. Bull.* **1978**, *26*, 2657.