A Simple Synthesis of 3′-*O*-(4-methoxytetrahydropyran-4-yl)-N^6-anisoyladenosine

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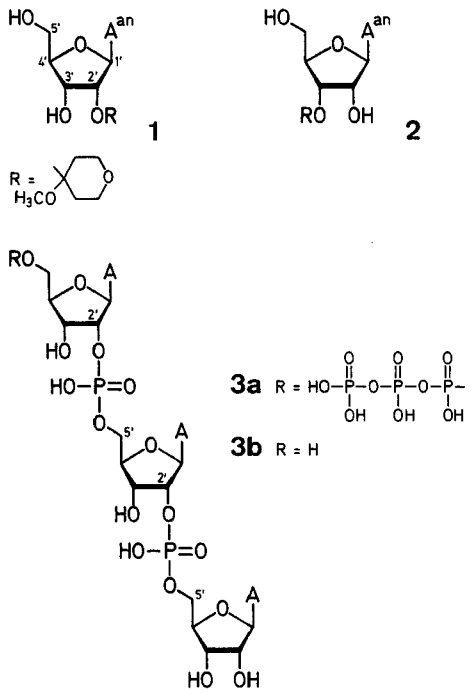
As part of our research on the synthesis of naturally occurring RNA fragments via phosphotriester intermediates, we devoloped[1] a general procedure for the synthesis of key-intermediate 2′-*O*-(4-methoxytetrahydropyran-4-yl) derivatives (2′-*O*-Mthp derivatives, e.g. **1**) of all four common ribonucleosides.

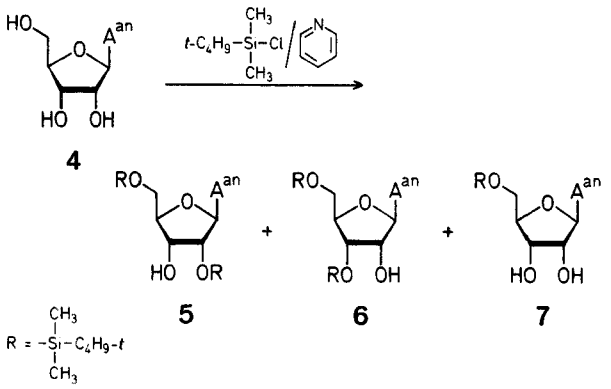
It has recently been shown[2-5] that trimers **3a, b**, which contain solely 2′-5′-internucleotide phosphodiester linkages, instead of 3′-5′-linkages, function as effective inhibitors of protein synthesis in eukaryotic cells. We now present a simple preparation of 3′-*O*-Mthp-N^6-anisoyladenosine **(2)** which may serve as an important building block for the synthesis of trimers **3a, b**.



The first step in our synthesis is based upon the use of *t*-butyldimethylsilyl (Tbdms) as a protective group for hydroxy functions. This protective group, which has been introduced by Corey et al.[6] in the prostoglandin field, has recently been successfully applied by Ogilvie et al.[7] in the preparation of di- and monosilylated ribonucleosides.

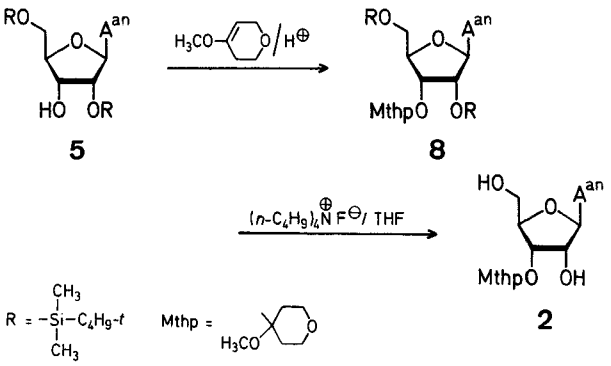
Treatment of N^6-anisoyladenosine[8] **(4)** with *t*-butyldimethylsilyl chloride[9] (3 mol equiv) in dry pyridine for 16 h at 50° gives a mixture of the silylated *N*-anisoyladenosine derivatives **5**, **6**, and **7** in the ratio 4:4:1.

The silylated products **5** and **6** can, due to their different chromatographic behaviour, be isolated after separation by thick-layer or short column chromatography. However, we found that crystallization of the concentrated crude reaction mixture, containing **5**, **6**, and **7**, from dry acetonitrile



affords pure **5**, and addition of water (1:3, v/v) to the mother liquor, which is now relatively rich in silylated derivative **6**, precipitates crystalline **6**. This selective crystallization procedure enables us to isolate, without using chromatographic methods, both silylated isomers **5** and **6** in satisfactory yields (35% and 25%, respectively).

The desired key derivative **2** of adenosine was easily obtained as a crystalline product in 80% yield by reaction of the 2′,5′-di-*O*-silyl derivative **5** with 4-methoxy-5,6-dihydro-2*H*-pyran[10] in dry dioxan in the presence of mesitylenesulfonic acid, treatment of the resultant crude, fully protected nucleoside **8** with tetrabutylammonium fluoride[7] in tetrahydrofuran, and recrystallization of the concentrated reaction mixture from water.



Using the same method, we prepared the isomeric 2′-*O*-Mthp derivative **1** starting from 3′,5′-di-*O*-silyl derivative **6**. Compound **1** thus obtained was identical (m.p., T.L.C., ^{1}H-N.M.R.) with an authentic specimen prepared from 3′-*O*-propanoyl-5′-*O*-methoxyacetyl-N^6-anisoyladenosine[1]. This experiment shows that the synthetic sequence leading to compound **1** (and as a consequence, also that leading to compound **2**) proceeds without any detectable isomerization of the silyl protective groups. Further, the difference between the R_f values of **2** and **1** (0.68 and 0.61, respectively, by T.L.C. on silica gel using chloroform/methanol 80:20 as eluent) may be used to check the (position-) isomeric purity of compounds **2** and **1**.

T.L.C. analyses were carried out on silica gel (TLC-Ready Plastic Sheets F 1500 LS 254 Silica Gel, Schleicher & Schüll). The ^{1}H-N.M.R. spectra were recorded on a Jeol JNM PS 100 spectrometer at 100 MHz

2′,5′- and 3′,5′-Bis[*O*-t-Butyldimethylsilyl]-N^6-anisoyladenosine (2′,5′- and 3′,5′-di-*O*-Tbdms-N^6-anisoyladenosine; 5, 6):
To a stirred solution of N^6-anisoyladenosine (7.26 g, 18.1 mmol) in dry pyridine (36 ml) is added commercial (Aldrich) *t*-butyldime-

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thylsilyl chloride (8.18 g, 54.3 mmol) and the mixture is left at 50°. T.L.C. analysis (chloroform/methanol, 97:3 v/v) after 16 h shows the following distribution of products: **5** (45%), **6** (40%), **7** (10%), and a trisilylated adenosine derivative (5%). Ice (25 g) is then added, the mixture is concentrated, and the residual oil is dissolved in chloroform (200 ml). The solution is washed with aqueous sodium hydrogen carbonate (10% w/v, 150 ml) and water (100 ml), dried with magnesium sulfate, and concentrated to an oil. Repeated co-evaporation with absolute ethanol (3 × 100 ml) gives a glass which is crystallized from hot anhydrous acetonitrile (150 ml) to give pure **5** (the mother liquor is saved); yield: 4.0 g (35%); m.p. 131°; R_f (chloroform/methanol 97:3): 0.52.

$C_{30}H_{47}N_5O_6Si_2$	calc.	C 57.24	H 7.47	N 11.12
(629.5)	found	56.37	7.60	10.80

U.V. (95% ethanol): $\lambda_{max} = 289$ ($\log \varepsilon = 4.52$), $\lambda_{min} = 239$ nm (3.72).

^{1}H-N.M.R. ($CDCl_3$/TMS): $\delta = 6.16$ ppm (d, 1H, 1′-H, $J_{1',2'} = 5.3$ Hz).

The pure product **6** is obtained by addition of water (50 ml) to the mother liquor of the isolation of **5**; yield: 2.85 g (25%); m.p. 83°; R_f (chloroform/methanol 97:3): 0.44.

$C_{30}H_{47}N_5O_6Si_2$	calc.	C 57.24	H 7.47	N 11.12
(629.5)	found	56.97	7.62	10.74

U.V. (95% ethanol): $\lambda_{max} = 288$ ($\log \varepsilon = 4.49$), $\lambda_{min} = 237$ nm (3.86).

^{1}H-N.M.R. ($CDCl_3$/TMS): $\delta = 6.06$ ppm (d, 1H, 1′-H, $J_{1',2'} = 4.1$ Hz).

3′-*O*-(4-Methoxytetrahydropyran-4-yl)-*N*^{6}-anisoyladenosine (2):
To a stirred solution of compound **5** (4.0 g, 6.35 mmol) in dry dioxan (24 ml) is added commercial (Aldrich) 4-methoxy-5,6-dihydro-2*H*-pyran (6 ml) and mesitylenesulfonic acid (300 mg, 1.5 mmol) and the mixture is left at 20° for 2 h. A second portion of 4-methoxy-5,6-dihydro-2*H*-pyran (6 ml) is added and after another 2 h T.L.C. analysis (chloroform/methanol, 97:3, v/v) shows the reaction to be complete. The mixture is neutralized with methanolic ammonia (half saturated at 0°) and concentrated under reduced pressure. The resultant oil is dissolved in chloroform (200 ml), washed with aqueous sodium hydrogen carbonate (10% w/v, 150 ml) and water (100 ml), dried with magnesium sulfate, and concentrated to an oil. A 0.5 molar solution of tetrabutylammonium fluoride in dry tetrahydrofuran (44.5 ml, 3.5 equiv) is added and after 1 h, T.L.C. analysis (chloroform/methanol, 80:20, v/v) shows the complete removal of the silyl groups. The mixture is concentrated, dissolved in chloroform (200 ml), and washed with aqueous sodium hydrogen carbonate (10% w/v, 100 ml) and water (100 ml). The organic layer is dried with magnesium sulfate, concentrated to an oil, and triturated with petroleum ether (40–60°, 2 × 250 ml). The precipitate is redissolved in chloroform and the solution evaporated under reduced pressure to give a glass. The latter is crystallized from boiling water (pH 7, 200 ml) to give pure **2**; yield 2.6 g (80% based on **5**); m.p. 123°; R_f (chloroform/methanol 80:20): 0.68.

$C_{24}H_{29}N_5O_8$	calc.	C 55.94	H 5.63	N 13.59
(515.2)	found	55.88	6.23	13.54

U.V. (95% ethanol): $\lambda_{max} = 290$ ($\log \varepsilon = 4.47$), $\lambda_{min} = 240$ nm (3.72).

^{1}H-N.M.R. (DMSO-d_6/D_2O/TMS): $\delta = 6.09$ ppm (d, 1H, 1′-H, $J_{1',2'} = 5.4$ Hz).

2′-*O*-(4-Methoxytetrahydropyran-4-yl)-*N*^{6}-anisoyladenosine (1):
This compound is obtained from **6** (2.4 g, 3.81 mmol) by a procedure analogous to that used for the synthesis of **2**; yield of pure **1**: 1.37 g (70% based on **6**); m.p. 135°; R_f (chloroform/methanol 80:20): 0.61.

$C_{24}H_{29}N_5O_8$	calc.	C 55.94	H 5.63	N 13.59
(515.2)	found	55.50	5.82	13.40

U.V. (95% ethanol): $\lambda_{max} = 289$ ($\log \varepsilon = 4.45$), $\lambda_{min} = 241$ nm (3.75).

^{1}H-N.M.R. (DMSO-d_6/D_2O/TMS): $\delta = 6.25$ ppm (d, 1H, 1′-H, $J_{1',2'} = 7.5$ Hz).

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