Synthesis of (2′*S*)-2′-Deoxy-2′-*C*-methylpurine Nucleosides and Their Phosphoramidites

by **Gerardo Caballero**[a][1]), **Mariana Gallo**[b]), **Javier M. Montserrat**[b]), and **Adolfo M. Iribarren***[b])[c])

[a]) Instituto di Ricerche di Biologia Molecolare (RBM), Roma, Italy
[b]) Instituto de Investigación en Ingeniería Genética y Biología Molecular (INGEBI), CONICET, Vuelta de Obligado 24902°P, (1428) Buenos Aires, Argentina (e-mail: airi@dna.uba.ar)
[c]) Universidad Nacional de Quilmes, Bernal, Argentina

We describe the stereoselective synthesis of (2′*S*)-2′-deoxy-2′-*C*-methyladenosine (**12**) and (2′*S*)-2′-deoxy-2′-*C*-methylinosine (**14**) as well as their corresponding cyanoethyl phosphoramidites **16** and **19** from 6-*O*-(2,6-dichlorophenyl)inosine as starting material. The methyl group at the 2′-position was introduced *via* a *Wittig* reaction (→ **3**, *Scheme 1*) followed by a stereoselective oxidation with OsO_4 (→ **4**, *Scheme 2*). The primary-alcohol moiety of **4** was tosylated (→ **5**) and regioselectively reduced with $NaBH_4$ (→ **6**). Subsequent reduction of the 2′-alcohol moiety with Bu_3SnH yielded stereoselectively the corresponding (2′*S*)-2′-deoxy-2′-*C*-methylnucleoside (→ **8a**).

1. Introduction. – The interest shown in the development of new synthetic nucleosides, with modifications at the base, at the sugar, or at both moieties, has been prompted by their biological activity as antiviral and anticancer agents. In recent years, a huge number of nucleoside analogues have been reported. Among them, those with modifications at the 2′-position of the sugar ring have found important medicinal applications (*e.g.*, araA [1], araC [2], DMDC [3], and SMIU [4]). In particular, it has been found that 2′-deoxy-2′-*C*-methylpyrimidine nucleosides with a (2′*S*)-configuration were much more effective in their biological activity than those that have (2′*R*)-configuration [3].

Another interesting application of nucleoside analogues is the synthesis of modified oligonucleotides due to their potential use in antisense strategy. For this particular approach, the modification should provide enhanced resistance to nucleases and hybridization affinity. Again, 2′-analogues demonstrated to be effective [5][6]. For example, 2′-*O*-alkyloligoribonucleotides [7][8] show increased nuclease resistance and form stable duplexes and 2′-*O*-methoxyethyloligonucleotides [9] seem to be very promising. The hybridization properties of some (2′*S*)-2′-deoxy-2′-*C*-methyloligonucleotides with pyrimidine bases [10][11] and their enhanced stability towards purified nucleases, human serum, and human cellular extracts have been reported [12]. Therefore, the preparation of the corresponding purine phosphoramidites seems to be a desirable objective.

We have previously developed a stereoselective synthesis for (2′*S*)-2′-deoxy-2′-*C*-methylpyrimidines [13]. This strategy implied the oxidation at the 2′-position, followed by a *Wittig* reaction at the resulting carbonyl group and the subsequent catalytic hydrogenation of the C=C bond. Unfortunately, this method was not appropriate for purine derivatives.

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[1]) Present address: Laboratorio de Control de Doping, Secretaría de Deporte y Recreación, Buenos Aires, Argentina.

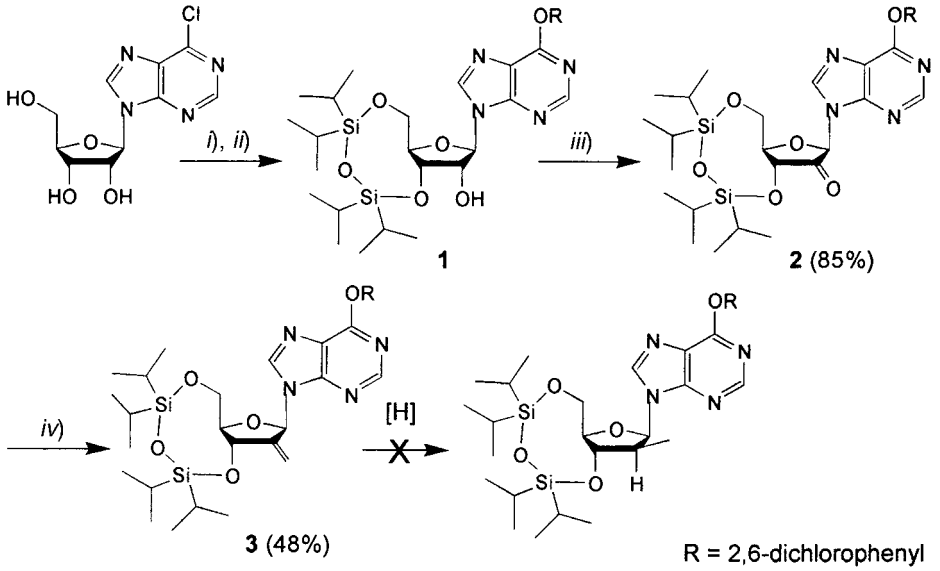
Previously reported synthesis of 2′-*C*-methyl analogues could be classified into two groups. In the first one, D-glucose is conveniently transformed into the corresponding 2′-*C*-methylribose; then a glycosylation step with the appropriate base affords the riboside analogue. This strategy is usually lengthy and affords low yields [14]. The second type of preparation involves the addition of organometallic reagents to the 2′-keto derivative of the nucleoside, followed by deoxygenation of the resulting alcohol [15]. However, since the reaction proceeds from the α-side of the riboside, the main reaction product has (2′*R*)-configuration.

Based on these facts, we decided to develop an alternative synthetic route for (2′*S*)-2′-deoxy-2′-*C*-methylpurine nucleosides based on the strategy recently published for (2′*S*)-2′-*C*-methylpuridine [16]. In this case, we initiated the synthesis with the versatile 6-*O*-(2,6-dichlorophenyl)inosine, which provided a common intermediate for the preparation of adenosine and inosine derivatives.

We also report the preparation of the 2-cyanoethyl phosphoramidites of (2′*S*)-2′-deoxy-2′-*C*-methylinosine and (2′*S*)-2′-deoxy-2′-*C*-methyladenosine suitable for solid-phase synthesis of modified oligonucleotides.

2. Results and Discussion. – 2.1. *3′,5′,6-*O*-Protected 2′-Deoxy-2′-methylideneinosine* **3**. The basic priority was to prepare (2′*S*)-2′-deoxy-2′-*C*-methylpurine ribosides without involving a glycosylation step and in a stereoselective way. The initial intention was to synthesize a 6-O-substituted 2′-deoxy-2′–methylideneinosine to obtain 2′-deoxy-2′-*C*-methyladenosine and 2′-deoxy-2′-*C*-methylinosine by hydrogenation of a common 2′-methylidere intermediate, as shown in *Scheme 1*.

Scheme 1. *Synthesis of 2′-Deoxy-2′-methylidenepurine Riboside* **3**

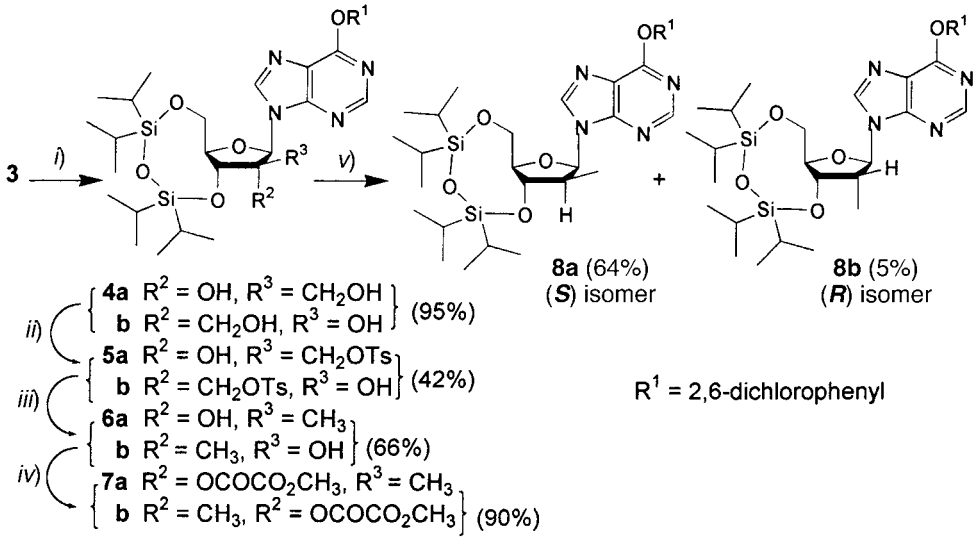


i) $TIPDSDCl_2$, Py. *ii*) 1,4-Diazabicyclo[2.2.2]octane (DABCO); Et_3N, 1,2-dichloroethane, 2,6-dichlorophenol. *iii*) CrO_3, Py, Ac_2O, CH_2Cl_2. *iv*) $Ph_3P{=}CH_2$, THF. [H]: H_2 (4 bar), AcOEt, 48 h.

Thus, 3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)-6-chloropurine riboside was prepared by using *Markiewicz*'s 1,3-dichloro-1,1,33-tetraisopropyldisiloxane ($TIPDSCl_2$) reagent [17] and 6-chloropurine riboside as starting materials. This compound was then converted to the more versatile 6-O-(2,6-dichlorophenyl)-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)inosine **1** by treatment with 2,6-dichlorophenol as previously reported [18]. The 2′-keto riboside **2** was obtained in 85% yield by oxidation with chromium oxide in pyridine and Ac_2O [19]. *Wittig* olefination of **2** with methyltriphenylphosphonium bromide in THF and in the presence of BuLi as base gave the 2′-methylenepurine nucleoside **3** in 48% yield.

2.2. *3′,5′,6*-O-*Protected (2′*S*)-2′-Deoxy-2′*-C-*methylinosine* **8a**. At this point, we tried to obtain the 2′-deoxy-2′-*C*-methylpurine derivative by simple hydrogenation of **3** with Pd/C as catalyst. Although different experimental conditions were tried, this reaction did not proceed. This finding can not be explained in terms of steric hindrance because the pyrimidine analogues were reduced in quantitative yields [13]. These results prompted us to change the strategy. We decided to apply the basic approach used in the synthesis of 2′-*C*-methyluridine [16], as outlined in *Scheme 2*.

Scheme 2. *Preparation of Intermediate* **8a**



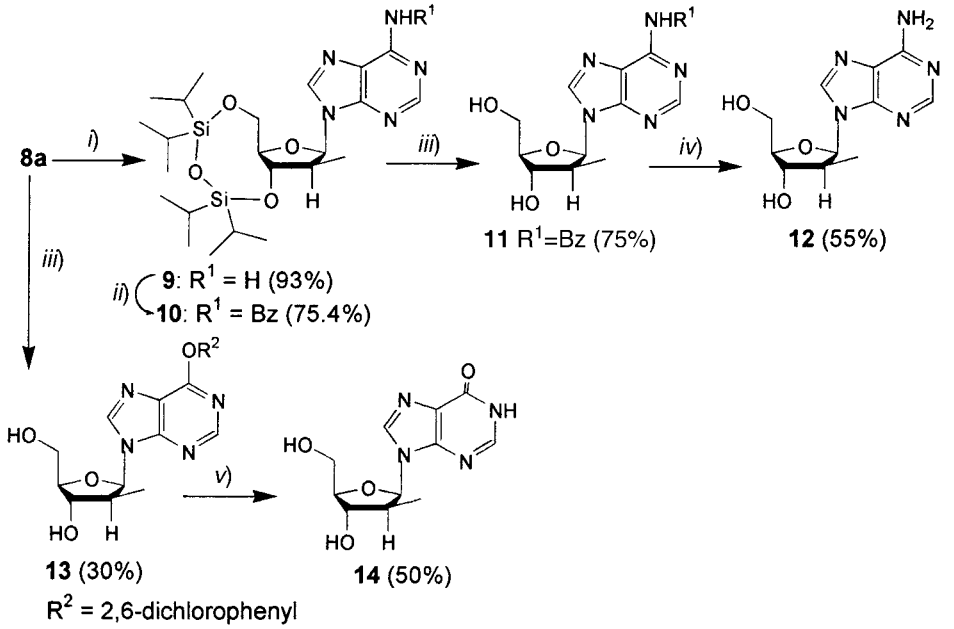
i) OsO_4, NMO, THF, *t*-BuOH, H_2O. *ii*) TsCl, Py. *iii*) $NaBH_4$, DMF. *iv*) $ClCOCOOCH_3$, DMAP, Py. *v*) Bu_3SnH, AIBN, toluene.

The 2′-methylenepurine riboside **3** was oxidized quantitatively with OsO_4 as catalyst [20], producing a diastereoisomer mixture of the corresponding diols **4a** and **4b**, in a 10:1 ratio, as determined by NMR. The major product was the (2′*R*)-2′-C-(hydroxymethyl)*inosine*, which is consistent with the attack of the OsO_4 from the less hindered side of the molecule. The mixture of diastereoisomers was then selectively tosylated at the primary-alcohol moiety, giving compouds **5a** and **5b** in 63% total yield. The mixture **5a**/**5b** was reduced with $NaBH_4$ in DMF at room temperature and the crude product purified by prep. HPLC (silica gel), yielding **6a**/**6b** in a total yield of 66%

and a diastereoisomer ratio equal to the one reported above for **4a**/**4b**. Reaction with methoxalyl chloride (=chlorooxoacetic acid methyl ester) in dry pyridine in the presence of *N*,*N*-dimethylpyridin-4-amine (DMAP) furnished the 2′-epimer mixture **7a**/**7b** in 90% total yield. The crude product was reduced without further purification by Bu_3SnH in dried toluene and AIBN (2,2′-azobis[2-methylpropanenitrile]) as a radical initiator. The final mixture was separated by HPLC, and diastereoisomers **8a** and **8b** were obtained in 64 and 5% yield, respectively.

2.3. *(2′S)-2′-Deoxy-2′-C-methylpurine Ribosides* **12** *and* **14**. To test whether the 2′-methylpurine ribosides have any biological activity, we prepared (2′*S*)-2′-deoxy-2′-*C*-methyladenosine (**12**) and (2′*S*)-2′-methyl-2′-*C*-deoxyinosine (**14**) from the common intermediate **8a** *via* the reactions depicted in *Scheme 3*. Treatment of **8a** with an ammonia solution in THF at 90° under pressure gave compound **9**. We decided to transform crude **9** into the benzoyl derivative **10**, which is a common intermediate in the preparation of the phosphoramidite and the free nucleoside and simplifies the purification step after removal of the disiloxane bridge. By this way, compound **11** was obtained in 75% yield, and final deprotection with aqueous ammonia led to pure **12**. To obtain (2′*S*)-2′-deoxy-2′-*C*-methylinosine, **8a** was desilylated under the usual conditions with Bu_4NF and the product purified by column chromatography (CC; silica gel) to give **13** in 60% yield. Treatment of **13** with 2-nitrobenzaldehyde oxime and tetramethylguanidine in MeCN [18] furnished (2′*S*)-2′-deoxy-2′-*C*-methylinosine (**14**) in 50% yield.

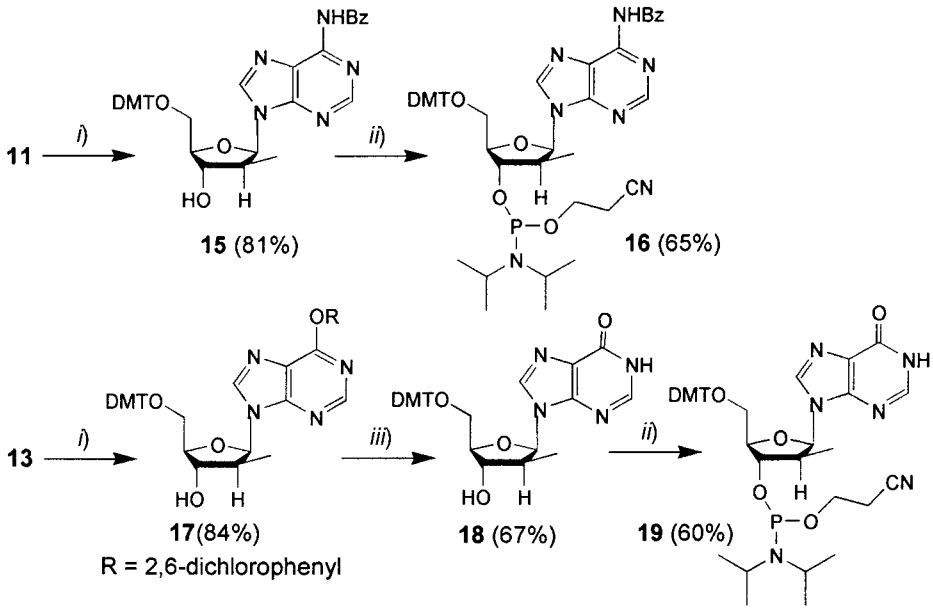
Scheme 3. *Synthesis of 2′-Deoxy-2′-C-methylpurine Ribosides* **12** *and* **14**



i) NH_3, THF. *ii*) BzCl, Py. *iii*) TBAF, THF. *iv*) NH_3(aq). *v*) 2-Nitrobenzaldehyde oxime, tetramethylguanidine, MeCN.

2.4. *(2′S)-2′-Deoxy-2′-C-methylpurine Nucleoside Phosphoramidites* **16** *and* **19**. We were also interested in the applications of (2′*S*)-2′-deoxy-2′-*C*-methylnucleosides to oligonucleotide synthesis. We have previously reported that pyrimidine oligonucleotides carrying this chemical modification showed enhanced resistance to degradation by nucleases [12]. Therefore, we synthesized the corresponding inosine and adenine phosphoramidites, as described in *Scheme 4*.

Scheme 4. *Synthesis of Phosphoramidites* **16** *and* **19**



i) $(MeO)_2Tr$, Et_3N, Py. *ii*) $iPr_2NPCl(OCH_2CH_2CN)$, iPr_2EtN, 1,2-dichloroethane. *iii*) 2-Nitrobenzaldehyde oxime, tetramethylguanidine, MeCN.

Starting from (2′*S*)-N^6-benzoyl-2′-deoxy-2′-*C*-methyladenosine (**11**), the 5′-position was protected as the dimethoxytrityl derivative following standard protocols [21a] to give **15** in 81% yield after purification by CC. The phosphoramidite **16** was obtained by reaction with 2-cyanoethyl diisopropylphosphoramidochloridite and $^iPr_2NEt$ [21b] as pure compound in 65% yield. The (2′*S*)-2′-deoxy-2′-*C*-methylinosine phosphoramidite **19** was prepared from compound **13**, which was first transformed into the dimethoxytrityl derivative **17** to simplify the purification step by enhancing the compound's lipophilicity. Treatment of this intermediate with 2-nitrobenzaldehyde oxime gave the inosine derivative **18**, which was purified by CC (silica gel) and isolated in 67% yield. Phosphitylation under standard conditions and CC purification afforded phosphoramidite **19**, which was fully characterized.

3. Conclusions. – The new modified (2′*S*)-2′-deoxy-2′-*C*-methylinosine and (2′*S*)-2′-deoxy-2′-*C*-methyladenosine as well as the corresponding 2-cyanoethyl phosphoramidite building blocks for solid-phase oligonucleotide synthesis were prepared.

Since the hydrogenation of the 2′-methylenepurine derivative was unsuccessful, the 2′-deoxy-2′-*C*-methylnucleosides were obtained by a strategy that was stereoselective in terms of the desired configuration.

The synthesis of oligonucleotides carrying these purine nucleoside analogues as well as structural and thermochemical analysis of the corresponding duplexes will be carried out in the near future. The biological activity of the modified nucleosides will also be assessed.

Experimental Part

General. AcOEt, toluene, dioxane,1,2-dichloroethane tetrahydrofuran (THF), MeOH, petroleum ether, Et_3N, pyridine (Py), dimethylformamide (DMF), ^{t}BuOH, Et_2O, HPLC-grade CH_2Cl_2, and MeCN, were supplied by *J. T. Baker*. THF was dried by refluxing over Na metal in the presence of benzophenone as an indicator of dryness and then distilled at atmospheric pressure. DMF was dried by heating at 100° with CaH_2 and then distilled under reduced pressure. Pyridine, dioxane, and Et_3N were dried by heating under reflux over CaH_2 and distilled at atmospheric pressure. All other reagents were commercially available (*Fluka*, *Aldrich*), and 6-chloropurine riboside was of the best anal. grade (*Pharma Waldhof*). All moisture-sensitive reagents were transferred *via* syringe under positive N_2 pressure or Ar atmosphere; moisture-sensitive reactions were carried out under positive pressure of N_2. In the case of mixtures, only the major product is described. Column chromatography (CC): silica gel 20–45 μm (*Amicon*). Anal. TLC: precoated *Merck* silica gel *60* F_{254} plates; detection by UV light, dimethoxytrityl products as orange spots after treatment by H_2SO_4/EtOH 8:2 and heating. Prep. HPLC: silica-gel cartridges, *Waters System-500-A* liquid chromatograph (*Waters Millipore*, Milford, USA); compounds **12** and **14** were purified by prep. reversed-phase HPLC by means of a *Kromasil* C_{18} (5 μm 30 × 1.0) column and *Gilson* equipment. NMR Spectra: $CDCl_3$ or (D_6)pyridine (*Aldrich*) solns.; 200-MHz *AM-Bruker* equipment; chemical shifts δ in ppm rel. to $SiMe_4$ (0.00 ppm, ^{1}H) and $CDCl_3$ (76.95 ppm, ^{13}C) as internal standards and 85% H_3PO_4 (0.00 ppm, ^{31}P) as external standard; δ of *m* from the approximated center, selected ^{13}C-NMR data are given in the *Table*. Microanalysis: recrystallized samples; automatic analyzer (*Fisons Instruments*).

*6-*O*-(2,6-Dichlorophenyl)-3′,5′-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)inosine* (**1**) was synthesized from 6-chloro-purine riboside as previously reported [16][17].

*2′-Deoxy-6-*O*-(2,6-dichlorophenyl)-2′-oxo-3′,5′-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)inosine* (**2**). Ac_2O (3.2 ml, 2.7 equiv.) and pyridine (5.4 ml, 6.5 equiv.) were added dropwise to an ice-cold suspension of CrO_3 (3.2 g, 3.0 equiv.) in CH_2Cl_2 (0.24 l). After the mixture became homogeneous (20 min), a soln. of 6-*O*-(2,6-dichlorophenyl)-3′,5′-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)inosine (7.1 g, 10.8 mmol) was added, and stirring continued for 1 h. The mixture was poured into cold AcOEt (1.4 l) and filtered through silica gel *60*. The filtrate was evaporated: crude **2** (6.0 g, 85%) as a foam. An anal. sample was obtained by CC petroleum ether/AcOEt 80:20. ^{1}H-NMR ($CDCl_3$): 0.80–1.20 (*m*, Me_2CH); 4.10 (*m*, H–C(4′)); 4.25 (*m*, 2 H–C(5′)); 5.55 (*m*, H–C(3′)); 5.68 (*d*, H–C(1′)); 7.30 (*m*, 2 H_m, H_p); 8.20 (*s*, H–C(2)); 8.40 (*s* H–C(8)). ^{13}C-NMR ($CDCl_3$): 12.4, 12.5, 12.9, 13.4 (Me_2CH); 16.7–17.3 (Me_2CH); 60.9 (C(5′)); 72.5 (C(3′)); 79.1 (C(4′)); 80.7 (C(1′)); 121.5 (C(5)); 127.3 (C_p); 128.8 (C_m); 129.5 (C_o); 143.0 (C(8)); 145.3 (C_{ipso}); 152.2, 152.5 (C(2), C(4)); 158.6 (C(6)); 205.3 (C(2′)). Anal. calc. for $C_{28}H_{38}Cl_2N_4O_6Si_2$) (653.71): C 51.45, H 5.86, N 8.57; found: C 51.72, H 6.18, N 8.54.

*2′-Deoxy-6-*O*-(2,6-dichlorophenyl)-2′-methylidene-3′,5′-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)inosine* (**3**). To a suspension of methyltriphenylphosphonium bromide (8.68 g; 1.6 equiv.) in dry THF (150 ml), 1.6M BuLi in hexane (18.4 ml, 1.9 equiv.) were carefully added under Ar, and the mixture was stirred at r.t. for 1 h. Then, a soln. of **2** (10.0 g, 15.3 mmol) in dry THF (10 ml) was added and stirring continued for 2 h. The reaction was quenched with 1N aq. NH_4Cl soln. (120 ml), the mixture extracted with AcOEt (2 × 160 ml), the combined org. phase washed with H_2O, dried (Na_2SO_4), and evaporated, and the residue purified by prep. HPLC (petroleum ether/AcOEt 75:25): 4.8 g (48%) of pure **3**. Foam. ^{1}H-NMR ($CDCl_3$): 0.80–1.20 (*m*, Me_2CH); 3.85 (*m*, H–C(4′)); 4.15 (*m*, 2 H–C(5′)); 5.35 (*m*, H–C(3′)); 5.56 (*m*, $=CH_2$); 6.68 (*d*, H–C(1′)); 7.30 (*m*, 2 H_m, H_p); 8.18 (*s*, H–C(2)); 8.45 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 12.5, 12.7, 13.0, 13.5 (Me_2CH); 16.8–17.4 (Me_2CH); 61.3 (C(5′)); 71.3 (C(3′)); 83.1[2]) (C(4′)); 83.5[2]) (C(1′)); 112.1 ($=CH_2$); 121.5 (C(5)); 127.1 (C_p); 128.8

[2]) Assignments may be interchanged.

Table. *Selected ^{13}C-NMR Chemical Shifts* [ppm] *for Compounds* **2**–**10**, **13**, *and* **15**–**19** *in* $CDCl_3$ *and* **11**, **12**, *and* **14** *in (*D_5*)pyridine*

	C(1′)	C(2′)	C(3′)	C(4′)	C(5′)	C(2)	C(4)	C(5)	C(6)	C(8)
2	80.7	205.3	72.5	79.1	60.9	152.2[a])	152.5[a])	121.5	158.6	143.0
3	83.5[a])	147.1	71.3	83.1[a])	61.3	152.8	152.0	121.5	158.3	141.7
4a	90.8	81.3	70.1	82.4	61.0	151.5	152.4	121.6	158.4	143.1
5a	89.5	79.3	70.0	81.8	60.9	151.8	152.7	121.7	158.4	142.4
6a	91.0	79.5	74.5	82.2	61.3	151.9	152.8	121.6	158.4	142.0
7a	90.7[a])	88.4[a])	74.3	84.5	60.4	152.1	152.8	121.6	158.4	142.1
8a	86.1[a])	44.9	73.4	81.5[a])	60.7	151.7	153.0	121.2	158.3	142.3
8b	89.8	43.0	71.2	83.9	62.3	149.9	151.8	121.3	158.5	141.8
9	85.8	44.9	73.4	84.4	60.7	149.4	152.8	119.3	155.5	139.0
10	86.1	44.8	73.3	84.4	60.6	152.3	149.8[a])	122.9	151.4[a])	141.6
11	87.0[a])	49.0	73.2	86.8[a])	60.4	152.3[a])	151.6[a])	125.6	152.3[a])	143.4
12	87.2	47.2	73.5	85.7	60.1	152.6	150.5	121.9	154.8	141.4
13	87.1	45.6	72.3	85.1	59.6	151.9	152.7	120.4	158.1	143.2
14	85.7[a])	44.7	71.7	85.2[a])	59.2	147.7[a])	148.2[a])	123.1	160.5	138.0
15	86.5	45.4	75.3	83.6	63.1	152.4	151.3	122.9	149.5	141.8
16	86.6	44.8	75.4	83.7	63.1	152.1	151.2	122.8	149.5	141.9
17	86.6	45.3	75.9	83.4	63.2	151.7	152.9	121.1	158.3	142.3
18	86.3	45.9	74.6	83.6	63.0	144.8	148.1	126.8	158.3	138.7
19	87.0	45.2	74.9	83.4	62.8	144.6	148.3	126.5	158.1	138.4

[a]) Assignments may be interchanged.

(C_m); 129.4 (C_o); 141.7 (C(8)); 145.3 (C_{ipso}); 147.1 (C(2′)); 152.0 (C(4)); 152.8 (C(2)); 158.3 (C(6)). Anal. calc. for $C_{26}H_{40}Cl_2N_4O_5Si_2$ (615.71): C 53.45, H 6.19, N 8.60; found: C 53.70, H 6.23, N 8.55.

*(2′*R*)-6-*O*-(2,6-Dichlorophenyl)-2′-*C*-(hydroxymethyl)-3′,5′-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-inosine* (**4a**). Compound **3** (4.8 g, 7.4 mmol) was dissolved in a mixture of THF (18.5 ml), ^{t}BuOH (18.5 ml), H_2O (5.5 ml), and 4-methylmorpholine 4-oxide (NMO; 1.02 g, 1.2 equiv.). Then OsO_4 (0.74 ml of a 2.5 wt.-% soln. in ^{t}BuOH, 0.01 êquiv.) was added under ice cooling. The mixture was stirred for 5 days at 4° and then quenched with 1M aq. $NaHSO_3$ (100 ml) and extracted with AcOEt (2 × 50 ml). The org. layer was washed with H_2O, dried (Na_2SO_4), and evaporated: crude **4a**/**4b** 10:1 (by ^{13}C-NMR) (4.8 g, 95%), which was used without further purification for recording NMR spectra and for the next step. ^{1}H-NMR ($CDCl_3$): 0.80–1.20 (*m*, Me_2CH); 3.45 (*m*, CH_2OH); 3.55 (br. *s*, 1 OH); 3.92 (br. *s*, CH_2OH); 4.08–4.40 (*m*, H–C(4′), 2 H–C(5′)); 4.52 (*d*, *J*(3′,4′) = 7, H–C(3′)); 6.10 (*s*, H–C(1′)); 7.32 (*m*, 2 H_m, H_p); 8.43 (*s*, H–C(2)); 8.58 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 12.5, 12.8, 13.0, 13.4 (Me_2*C*H); 16.8–17.4 (*Me*$_2$CH); 61.0 (C(5′)); 62.5 (CH_2OH); 70.1 (C(3′)); 81.3 (C(2′)); 82.4 (C(4′)); 90.8 (C(1′)); 121.6 (C(5)); 127.1 (C_p); 128.7 (C_m); 129.3 (C_o); 143.1 (C(8)); 145.2 (C_{ipso}); 151.5 (C(2)); 152.4 (C(4)); 158.4 (C(6)). Anal. calc. for $C_{29}H_{42}Cl_2N_4O_7Si_2$ (685.75): C 50.79, H 6.17, N 8.17; found: C 50.47, H 6.12, N 8.11.

*(2′*R*)-6-*O*-(2,6-Dichlorophenyl)-2′-*C*-({[(4-methylphenyl)sulfonyl]oxy}methyl)-3′,5′-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)inosine* (**5a**). To a soln. of **4a**/**4b** (4.8 g, 7.0 mmol) in dry pyridine (95 ml), TsCl (6.5 g, 5 equiv.) was added and the mixture stirred overnight. Then, more TsCl (2.5 g; 3.5 equiv.) was added, and stirring was continued for 3 h. The solvent was evaporated, the residue dissolved in AcOEt, the soln. washed with H_2O, dried (Na_2SO_4), and evaporated, and crude product purified by filtration through silica gel *60* (CH_2Cl_2, then petroleum ether/AcOEt 1:1) **5a**/**5b** (3.74 g, 63%). ^{1}H-NMR ($CDCl_3$):0.80–1.20 (*m*, Me_2CH); 2.40 (*s*, Me (T_S)); 3.40 (br. *s*, OH); 3.88–4.35 (*m*, 5 H–C(4′), 2 H–C(5′), OCH_2–C(2′)); 4.85 (*d*, *J*(3′,4′) = 7, H–C(3′)); 6.10 (*s*, H–C(1′)); 7.32 (*m*, 2 H_m, H_p (Ar)); 8.25 (*s*, H–C(2)); 8.38 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 12.6, 12.7, 12.9, 13.6 (Me_2*C*H); 17.0–17.3 (*Me*$_2$CH); 21.6 (Me(Ts)); 60.9 (C(5′)); 66.5 (O*C*H_2–C(2′)); 70.0 (C(3′)); 79.3 (C(2′)); 81.8 (C(4′)); 89.5 (C(1′)); 121.7 (C(5)); 127.1 (C_p(Ar)); 127.9 (C_o(Ts)); 128.8 (C_m(Ar)); 129.5 (C_o(Ar)); 129.7 (C_m(Ts)); 131.6 (C_p(Ts)); 142.4 (C(8)); 145.2 (C_{ipso}(Ts)); 145.4 (C_{ipso}(Ar)); 151.8 (C(2)); 152.7 (C(4)); 158.4 (C(6)). Anal. calc. for $C_{36}H_{48}Cl_2N_4O_9SSi_2$ (839.94): C 51.48, H 5.76, N 6.67; found: C 51.24, H 5.69, N 6.62.

*(2′*R*)-6-*O*-(2,6-Dichlorophenyl)-2′-*C*-methyl-3′,5′-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)inosine* (**6a**). To a soln. of **5a**/**5b** (3.74 g, 4.4 mmol) in anh. DMF (46 ml), $NaBH_4$ (0.5 g, 3 equiv.) was added and the mixture kept at r.t. for 2 h. The reaction was quenched by addition of a sat. NH_4Cl soln., the mixture extracted with CH_2Cl_2 (2 × 50 ml), the combined org. phase washed with brine (5 × 100 ml), dried (Na_2SO_4), and evaporated, and the crude product (2.9 g) purified by prep. HPLC (petroleum ether/AcOEt 80:20): **6a**/**6b** (1.94 g, 66%). ^{1}H-NMR ($CDCl_3$): 0.80–1.20 (*m*, Me_2CH); 1.28 (*s*, Me–C(2′)); 3.15 (br. *s*, 1 OH); 4.05–4.38 (*m*, H–C(4′), 2 H–C(5′)); 4.62 (*d*, *J*(3′,4′) = 7, H–C(3′)); 6.10 (*s*, H–C(1′)); 7.30 (*m*, 2 H_m, H_p); 8.38 (*s*, H–C(2)); 8.50 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 12.7, 13.0, 13.6 (Me_2CH); 17.0–17.4 (Me_2CH); 21.0 (*Me*–C(2′)); 61.3 (C(5′)); 74.5 (C(3′)); 79.5 (C(2′)); 82.2 (C(4′)); 91.0 (C(1′)); 121.6 (C(5)); 127.1 (C_p); 128.8 (C_m); 129.5 (C_o); 142.0 (C(8)); 145.4 (C_{ipso}); 151.9 (C(2)); 152.8 (C(4)); 158.4 (C(6)). Anal. calc. for $C_{29}H_{42}Cl_2N_4O_6Si_2$ (669.76): C 52.01, H 8.37, N 6.32; found: C 52.34, H 8.42, N 6.28.

*(2′*R*)-6-*O*-(2,6-Dichlorophenyl)-2′-*C*-methyl-3′,5′-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)inosine 2′-(Methyl ethanedioate)* (**7a**). To a soln. of **6** (1.93 g, 2.9 mmol) and DMAP (72 mg, 0.2 equiv.) in anh. pyridine (51 ml), methoxalyl chloride (0.48 ml, 1.8 equiv.) was added under Ar, and the mixture was stirred for 1 h at r.t. The reaction was quenched with MeOH (3.5 ml), the solvent evaporated, the residue dissolved with AcOEt, and the soln. washed with H_2O, dried (Na_2SO_4), and evaporated: **7a**/**7b** (1.97 g, 90%), pure by TLC (toluene/acetone 90:10; R_f 0.4), which was used without further purification. An anal. sample for recording NMR spectra was obtained by CC. ^{1}H-NMR ($CDCl_3$): 0.80–1.20 (*m*, Me_2CH); 1.59 (*s*, Me–C(2′)); 3.95 (*s*, MeO); 4.08–4.38 (*m*, H–C(4′), 2 H–C(5′)); 4.78 (*d*, 1 *J*(3′,4′) = 8, H–C(3′)); 6.58 (*s*, H–C(1′)); 7.21 (*t*, *J*(*m*,*p*) = 9, H_p); 7.42 (*d*, *J*(*m*,*p*) = 9, 2 H_m); 8.45 (*s*, H–C(2)); 8.50 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 12.8, 12.9, 13.1 (Me_2CH); 13.6 (*Me*–C(2′)); 16.9–17.3 (Me_2CH); 53.4 (MeO); 60.4 (C(5′)); 74.3 (C(3′)); 81.5 (C(4′)); 88.4[2]) (C(2′)); 90.7[2]) (C(1′)); 121.6 (C(5)); 127.1 (C_p); 128.7 (C_m); 129.4 (C_o); 142.1 (C(8)); 145.3 (C_{ipso}); 152.1 (C(2)); 152.8 (C(4)); 156.2, 157.9 (O*C*O*C*OO); 158.4 (C(6)). Anal. calc. for $C_{29}H_{44}Cl_2N_4O_9Si_2$ (719.77): C 50.85, H 5.87, N 7.41; found: C 50.94, H 5.90, N 7.49.

*(2′*S*)-2′-Deoxy-6-*O*-(2,6-dichlorophenyl)-2′-*C*-methyl-3′,5′-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)inosine* (**8a**). The soln. of **7a**/**7b** (1.82 g, 2.4 mmol) in anh. toluene (22 ml) was carefully deoxygenated by Ar bubbling and removing gases by means of vacuum. This procedure was repeated 3 times, and then the soln. was heated at 100°. AIBN (0.22 g, 0.6 equiv.) and tributylstannane (1.3 ml, 2 equiv.) were added. The mixture was refluxed for 2 h, concentrated to half the volume *in vacuo*, and then purified by prep. HPLC (petroleum ether/AcOEt 85:15). The last traces of tributylhydroxystannane were eliminated dissolving the fractions obtained by HPLC in MeCN and washing this soln. 5 times with petroleum ether: **8a** (1.01 g, 64%) and **8b** ((2′*R*)-epimer; 0.1 g, 6%).

Data of **8a**: ^{1}H-NMR ($CDCl_3$): 0.85 (*d*, *J*(2′,Me) = 7, Me–C(2′)); 0.90–1.25 (*m*, Me_2CH); 2.82 (*m*, H–C(2′)); 3.90 (*m*, H–C(4′)); 4.08 (*dd*, *J*(5′a,4′) = 2, J_{gem} = 12, H_a–C(5′)); 4.27 (*dd*, J_{gem} = 12, *J*(4′,5′b) = 1, H_b–C(5′)); 4.41 (*dd*, *J*(3′,2′) = *J*(3′,4′) = 7, H–C(3′)); 6.48 (*d*, *J*(1′,2′) = 7, H–C(1′)); 7.20 (*t*, *J*(*m*,*p*) = 9, H_p); 7.42 (*d*, *J*(*m*,*p*) = 9, 2 H_m); 8.45 (*s*, H–C(2)); 8.52 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 11.2 (*Me*–C(2′)); 12.8, 12.9, 13.1, 13.6 (Me_2CH); 16.9–17.5 (Me_2CH); 44.9 (C(2′)); 60.7 (C(5′)); 73.4 (C(3′)); 84.5[2]) (C(4′)); 86.1[2]) (C(1′)); 121.2 (C(5)); 127.0 (C_p); 128.7 (C_m); 129.4 (C_o); 142.3 (C(8)); 148.5 (C_{ipso}); 151.7 (C(2)); 153.0 (C(4)); 158.3 (C(6)). Anal. calc. for $C_{29}H_{42}Cl_2N_4O_5Si_2$ (635.76): C 53.28, H 6.48, N 8.57; found: C 53.48, H 6.52, N 8.55.

Data of **8b**: ^{1}H-NMR ($CDCl_3$): 1.00 (*d*, *J*(2′,Me) = 7, Me–C(2′)); 0.90–1.25 (*m*, Me_2CH); 2.90 (*m*, H–C(2′)); 3.88–4.20 (*m*, H–C(4′), 2 H–C(5′)); 4.90 (*m*, H–C(3′)); 5.98 (*d*, *J*(1′,2′) = 7, H–C(1′)); 7.20 (*t*, *J*(*m*,*p*) = 9, H_p); 7.45 (*d*, *J*(*m*,*p*) = 8, 2 H_m); 8.45 (*s*, H–C(2)); 8.52 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 10.9 (*Me*–C(2′)); 12.7, 12.8, 13.0, 13.4 (Me_2CH); 17.0–17.5 (Me_2CH); 43.0 (C(2′)); 62.3 (C(5′)); 71.2 (C(3′)); 83.9 (C(4′)); 89.8 (C(1′)); 121.3 (C(5)); 127.7 (C_p); 128.8 (C_m); 129.5 (C_o); 141.8 (C(8)); 144.2 (C_{ipso}); 149.9 (C(2)); 151.8 (C(4)); 158.5 (C(6)). Anal. calc. for $C_{29}H_{42}Cl_2N_4O_5Si_2$ (635.76): C 53.28, H 6.48, N 8.57; found: C 53.57, H 6.50, N 8.53.

*(2′*S*)-2′-Deoxy-2′-*C*-methyl-3′,5′-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)adenosine* (**9**). A soln. of **8a** (0.37 g, 0.57 mmol) in anh. THF (15 ml) was treated for 5 days with aq. ammonia in a stainless steel bomb at 90°. Then, the solvent was evaporated, the residue dissolved in AcOEt, and the soln. washed with 5% aq. NaOH soln. and H_2O, dried (Na_2SO_4), and evaporated: **9** (267 mg, 93%). An anal. sample was purified by CC for recording NMR spectra. ^{1}H-NMR ($CDCl_3$): 0.80 (*d*, *J*(2′,Me) = 7, Me–C(2′)); 0.90–1.25 (*m*, Me_2CH); 2.80 (*m*, H–C(2′)); 3.88 (*m*, H–C(4′)); 4.05 (*d*, J_{gem} = 12, 1 H–C(5′)); 4.28 (*d*, J_{gem} = 12, 1 H–C(5′)); 4.41 (*dd*, *J*(3′,2′) = *J*(3′,4′) = 7, H–C(3′)); 5.62 (br. *s*, NH_2); 6.40 (*d*, *J*(1′,2′) = 7, H–C(1′)); 8.21 (*s*, H–C(2)); 8.35 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 11.2 (*Me*–C(2′)); 12.5, 12.9, 13.6 (Me_2CH); 16.9–17.4 (Me_2CH); 44.9 (C(2′)); 60.7 (C(5′)); 73.4 (C(3′)); 84.4 (C(4′)); 85.8 (C(1′)); 119.3 (C(5)); 139.0 (C(8)); 149.4 (C(4)); 152.8 (C(2)); 155.5 (C(6)). Anal. calc. for $C_{23}H_{41}N_5O_4Si_2$ (507.78): C 54.40, H 8.14, N 13.79; found: C 54.25, H 8.12, N 13.75.

(2′S)-N⁶-Benzoyl-2′-deoxy-2′-C-methyl-3′,5′-O-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)adenosine (**10**). To a soln. of **9** (238 mg, 0.47 mmol) in anh. pyridine (4.2 ml), benzoyl chloride (0.26 ml, 4.7 mmol) was added at 0°, and the mixture was stirred at r.t. for 3 h. The reaction was quenched by addition of H_2O (0.85 ml) followed by 15M NH_4OH (0.85 ml), and this mixture was kept at r.t. for 30 min. The solvents were evaporated, the oil thus formed was dissolved in Et_2O and the soln. washed with 1M $NaHCO_3$ and H_2O, dried (Na_2SO_4), and evaporated: 278 mg of crude **10**. An anal. sample was purified by CC for recording NMR spectra. ^{1}H-NMR ($CDCl_3$): 0.82 (*d*, *J*(2′,Me) = 7, Me–C(2′)); 0.90–1.25 (*m*, Me_2CH); 2.85 (*m*, H–C(2′)); 3.89 (*m*, H–C(4′)); 4.05 (*d*, J_{gem} = 12, 1 H–C(5′)); 4.28 (*d*, J_{gem} = 12, 1 H–C(5′)); 4.42 (*dd*, *J*(3′,2′) = *J*(3′,4′) = 8, H–C(3′)); 6.48 (*d*, *J*(1′,2′) = 6, H–C(1′)); 7.50 (*m*, 2 H_m, H_p); 8.10 (*d*, *J*(2,3) = 8, 2 H_o); 8.38 (*s*, H–C(2)); 8.75 (*s*, H–C(8)); 9.35 (br. *s*, HNCO). ^{13}C-NMR ($CDCl_3$): 11.2 (*Me*–C(2′)); 12.5, 12.9, 13.5 (Me_2CH); 16.9–17.4 (*Me*$_2$CH); 44.8 (C(2′)); 60.6 (C(5′)); 73.3 (C(3′)); 84.4 (C(4′)); 86.1 (C(1′)); 122.9 (C(5)); 128.1 (C_o); 129.8 (C_m); 131.8 (C_p); 133.9 (C_{ipso}); 141.6 (C(8)); 149.8[2]) (C(4)); 151.4[2]) (C(6)); 152.3 (C(2)); 165.3 (CONH). Anal. calc. for $C_{30}H_{45}N_5O_5Si_2$ (611.89): C 58.89, H 7.41, N 11.45; found: C 58.95, H 7.44, N 11.42.

(2′S)-N⁶-Benzoyl-2′-deoxy-2′-C-methyladenosine (**11**). A soln. of **10** (260 mg, 0.43 mmol) in anh. THF (1.2 ml) was treated with 1M Bu_4NF in THF (1.05 ml, 2.4 equiv.) at r.t. Five minutes later, the mixture was diluted with pyridine/MeOH/H_2O 3:3:1 (2.0 ml) and the soln. poured into a stirred suspension of *Dowex-50Wx4-200* resin (2.0 g; pyridinium form) in pyridine/MeOH/H_2O 3:3:1 (3.0 ml). The mixture was stirred for 30 min and the resin filtered off and washed with MeOH (3 × 5 ml). The combined filtrate and washings were evaporated, and the residual pyridine was removed by co-evaporation with toluene. The crude product (250 mg) was purified by CC (silica gel, CH_2Cl_2/MeOH 93:7): pure **11** (120 mg, 69% from **9**). ^{1}H-NMR ((D$_5$)pyridine): 0.91 (*d*, *J*(2′,Me) = 7, Me–C(2′)); 3.00 (*m*, H–C(2′)); 4.29–4.45 (*m*, H–C(4′), 2 H–C(5′)); 4.90 (*dd*, *J*(3′,2′) = *J*(3′,4′) = 7, H–C(3′)); 5.15 (br. *s*, 2 OH); 6.90 (*d*, *J*(1′,2′) = 7, H–C(1′)); 7.40 (*m*, 2 H_m, H_p); 8.30 (*d*, *J*(2,3) = 8, 2 H_o); 8.98 (*s*, H–C(2)); 9.55 (*s*, H–C(8)). ^{13}C-NMR ((D$_5$)pyridine): 11.5 (*Me*–C(2′)); 49.0 (C(2′)); 60.4 (C(5′)); 73.2 (C(3′)); 86.8[2]) (C(4′)); 87.0[2]) (C(1′)); 125.6 (C(5)); 128.8[2]) (C_o); 129.2[2]) (C_m); 132.4 (C_p); 135.9 (C_{ipso}); 143.4 (C(8)); 151.6[2]) (C(4)); 152.3[2]) (C(6)); 153.0[2]) (C(2)); 167.4 (CONH). Anal. calc. for $C_{18}H_{19}N_5O_4$ (369.38): C 58.53, H 5.18, N 18.96; found: C 58.32, H 5.22, N 18.84.

(2′S)-2′-Deoxy-2′-C-methyladenosine (**12**). Overnight, **11** (50 mg, 0.13 mmol) was kept stirring at 55° in aq. conc. ammonia. When the reaction was completed, the ammonia was evaporated and the residue dissolved in H_2O (20 ml) and washed 3 times with AcOEt. The aq. phase was evaporated and the residue purified by prep. reversed-phase HPLC (MeOH/H_2O 40:60): 55% **12**. ^{1}H-NMR ((D$_5$)pyridine): 0.70 (*d*, *J*(2′,Me) = 7, Me–C(2′)); 2.65 (*m*, H–C(2′)); 3.85–4.05 (*m*, H–C(4′), 2 H–C(5′)); 4.22 (*dd*, *J*(3′,2′) = *J*(3′,4′) = 7, H–C(3′)); 4.95 (br. *s*, 3 H, OH); 6.43 (*d*, *J*(1′,2′) = 7, H–C(1′)); 8.15 (*s*, H–C(2′)); 8.55 (*s*, H–C(8)). ^{13}C-NMR ((D$_5$)pyridine): 11.4 (*Me*–C(2′)); 47.2 (C(2′)); 60.1 (C(5′)); 73.5 (C(3′)); 85.7 (C(4′)); 87.2 (C(1′)); 121.9 (C(5)); 141.4 (C(8)); 150.5 (C(4)); 152.6 (C(2)); 154.8 (C(6)). Anal. calc. for $C_{11}H_{15}N_5O_3$ (265.27): C 49.80, H 5.70, N 26.40; found: C 49.93, H 5.74, N 26.57.

*(2′S)-2′-Deoxy-6-*O*-(2,6-dichlorophenyl)-2′-C-methylinosine* (**13**). Compound **8a** (569 mg, 0.87 mmol) was desilylated as described above for **11**. The mixture was purified by CC (CH_2Cl_2/MeOH 95:5): pure **13** (214 mg, 60%). ^{1}H-NMR ($CDCl_3$): 0.77 (*d*, *J*(2,Me) = 7, Me–C(2′)); 2.80 (*m*, H–C(2′)); 3.75 (br. *s*, 1 OH); 4.05 (*m*, 1 OH, 2 H–C(5′)); 4.33 (*m*, H–C(4′)); 4.46 (*m*, H–C(3′)); 6.50 (*d*, $J_{(1',2')}$ = 7, H–C(1′)); 7.32 (*m*, 2 H_m, H_p); 8.42 (*s*, H–C(2)); 9.01 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 11.2 (*Me*–C(2′)); 45.6 (C(2′)); 59.6 (C(5′)); 72.3 (C(3′)); 85.1 (C(4′)); 87.1 (C(1′)); 120.4 (C(5)); 127.3 (C_p); 128.8 (C_m); 129.3 (C_o); 143.2 (C(8)); 145.2 (C_{ipso}); 151.9 (C(2)); 152.7 (C(4)); 158.1 (C(6)). Anal. calc. for $C_{17}H_6Cl_2N_4O_4$ (411.25): C 49.65, H 3.92, N 13.62; found: C 49.42, H 3.87, N 13.70.

(2′S)-2′-Deoxy-2′-C-methylinosine (**14**). To a soln. of 2-nitrobenzaldehyde oxime (610 mg, 6.5 equiv.) and tetramethylguanidine (0.21 ml, 3 equiv.) in anh. MeCN (3 ml), **13** (150 mg, 0.36 mmol) was added, and the mixture was refluxed for 18 h and then evaporated. The residue was dissolved in CH_2Cl_2 and extracted with H_2O (twice). The aq. phase was washed with CH_2Cl_2 and evaporated to yield the crude product, which was purified by prep. reversed-phase HPLC (MeOH/H_2O 40:60); 50% **14**. ^{1}H-NMR ((D$_5$)pyridine): 0.73 (*d*, *J*(2′,Me) = 7, Me–C(2′)); 2.68 (*m*, H–C(2′)); 3.80–4.05 (*m*, H–C(4′), 2 H–C(5′)); 4.28 (*dd*, *J*(3′,2′) = *J*(3′,4′) = 7, H–C(3′)); 4.90 (br. *s*, 3 H, OH); 6.41 (*d*, *J*(1′,2′) = 7, H–C(1′)); 8.09 (*s*, H–C(2)); 8.51 (*s*, H–C(8)). ^{13}C-NMR ((D$_5$)pyridine): 10.6 (*Me*–C(2′)); 44.7 (C(2′)); 59.2 (C(5′)); 71.7 (C(3′)); 85.2[2]) (C(4′)); 85.7[2]) (C(1′)); 123.1 (C(5)); 138.0 (C(8)); 147.7[2]) (C(2)); 148.2[2]) (C(4)); 160.5 (C(6)). Anal. calc. for $C_{11}H_{14}N_4O_4$ (266.26): C 49.62, H 5.30, N 21.04; found: C 49.93, H 5.35, N 21.15.

*(2′S)-N⁶-Benzoyl-2′deoxy-5′-*O*-(4,4′-dimethoxytrityl)-2′-C-methyladenosine* (**15**). Compound **11** (86 mg, 0.23 mmol) was dried by evaporation of pyridine. Anh. pyridine (2.5 ml), Et_3N (0.06 ml, 1.9 equiv.), and 4,4′-dimethoxytrityl chloride (96 mg, 1.23 equiv.) were added with stirring and exclusion of moisture, and the

mixture was stirred at r.t. for 3 h. After this time, the reaction was incomplete; therefore, more 4,4′-dimethoxytrityl chloride (96 mg, 1.23 equiv.) and Et_3N (0.06 ml; 1.23 equiv.) were added, and stirring was continued for 24 h. The reaction was quenched with MeOH (0.5 ml), and the solvents were evaporated. The residue was dissolved in CH_2Cl_2, and the soln. was washed with 5% $NaHCO_3$ soln. and H_2O, dried (Na_2SO_4), and evaporated. Removal of the residual pyridine by co-evaporation with toluene afforded 293 mg of crude product, which was purified by CC (CH_2Cl_2/MeOH/Et_3N 97:2:1): pure **12** (127 mg, 81%). ^{1}H-NMR ($CDCl_3$): 0.78 (*d*, *J*(2′,Me) = 7, Me–C(2′)); 2.71 (*m*, H–C(2′)); 3.55 (*m*, 2 H–C(5′)); 3.78 (*s*, 2 MeO); 4.02 (*m*, H–C(4′)); 4.90 (*dd*, *J*(3′,2′) = *J*(3′,4′) = 8, H–C(3′)); 6.51 (*d*, *J*(1′,2′) = 7, H–C(1′)); 6.81 (*d*, *J* = 9, 4 H, $(MeO)_2Tr$); 7.23–7.61 (*m*, 8 H, $(MeO)_2Tr$, 2 H_m, H_p); 7.35 (*d*, *J* = 9, 4 H, $(MeO)_2Tr$); 8.30 (*d*, *J*(2,3) = 8, 2 H_o); 8.22 (*s*, H–C(2)); 9.55 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$):11.1 (*Me*–C(2′)); 45.4 (C(2′)); 55.1 ((*Me*O)$_2Tr$); 63.1 (C(5′)); 75.3 (C(3′)); 83.6 (C(4′)); 86.5[2]) (C(1′)); 86.8[2]) (*C*Ar$_3$ ((MeO)$_2Tr$)); 113.2 ((MeO)$_2Tr$); 122.9 (C(5)); 127.8, 127.9, 128.2, 128.8, 130.0, 132.6, 133.8, 135.6 ((MeO)$_2Tr$, *Ph*CO); 141.8 (C(8)); 144.3 ((MeO)$_2Tr$); 149.5 (C(6)); 151.3 (C(4)); 152.4 (C(2)); 158.6 ((MeO)$_2Tr$); 164.6 (Ph*C*O). Anal. calc. for $C_{39}H_{37}N_5O_6$ (671.75): C 69.73, H 5.55, N 10.43; found: C 69.94, H 5.59, N 10.48.

*(2′*S*)-*N^6*-Benzoyl-2′-deoxy-5′*-O-*(4,4′-dimethoxytrityl)-2′*-C-methyladenosine 3′-(2-cyanoethyl diisopropylphosphoramidite)* (**16**). Compound **15** (117 mg, 0.17 mmol) was dried by evaporation of MeCN. The foam was dissolved in dry 1,2-dichloroethane (0.8 ml) containing ^{i}Pr$_2$EtN (0.06 ml, 2 mmol) under Ar and the soln. was cooled in an ice-bath. Under stirring, 2-cyanoyl diisopropylphosphoramidochloridite (0.05 ml, 1.4 equiv.) was added dropwise within 2 min. The mixture was stirred at r.t. for 2 h and, since the reaction was not complete, more 2-cyanoethyl diisopropylphosphoramidochloridite (0.03 ml, 0.7 equiv.) was added and stirring continued for an additional hour. CH_2Cl_2 (10 ml) was added, the soln. washed with 5% aq. $NaHCO_3$ soln. and brine, dried (Na_2SO_4), and evaporated, and the residue purified by CC (CH_2Cl_2/MeOH/Et_3N 98:1:1): pure **16** (89 mg, 65%). ^{1}H-NMR ($CDCl_3$): 0.80 (*d*, *J*(2′,Me) = 7, Me–C(2′)); 1.18–1.50 (*m*, 2 *Me*$_2$CH); 2.70 (*m*, H–C(2′)); 2.81 (*m*, CH_2CH_2CN); 3.58 (*m*, C*H*$_2$CH$_2$CN, 2 H–C(5′)); 3.76 (*s*, 2 MeO); 3.85 (*m*, 2 Me$_2$C*H*); 4.01 (*m*, H–C(4′)); 4.35 (*m*, H–C(3′)); 6.58 (*d*, *J*(1′,2′) = 7, H–C(1′)); 6.80 (*d*, *J* = 9, 4 H, $(MeO)_2Tr$); 7.13–7.60 (*m*, 12 H, $(MeO)_2Tr$, 2 H_m, H_p; 7.92 (*d*, *J*(2,3) = 8, 2 H_o); 8.25 (*s*, H–C(2)); 8.62 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 11.2 (*Me*–C(2′)); 21.4 (CH_2CH_2CN); 24.4, 24.5, 24.6 (*Me*$_2$CH); 43.1, 43.3 (Me$_2$*C*H); 44.8 (C(2′)); 55.2 ((*Me*O)$_2Tr$); 57.4 (*C*H$_2$CH$_2$CN); 63.1 (C(5′)); 75.4 (C(3′)); 83.7 (C(4′)); 86.6[1]) (C(1′)); 86.8[1]) (*C*Ar$_3$ ((MeO)$_2Tr$)); 113.2 ((MeO)$_2Tr$); 117.3 (CN); 122.8 (C(5)); 127.0, 127.9, 128.3, 128.3, 128.8, 130.1, 132.7, 133.7, 135.7 ((MeO)$_2Tr$, *Ph*CO); 141.9 (C(8)); 144.4 ((MeO)$_2Tr$); 149.5 (C(6)); 151.2 (C(4)); 152.1 (C(2)); 158.6 ((MeO)$_2Tr$); 164.5 (Ph*C*O). ^{31}P-NMR ($CDCl_3$): +148.4. Anal. calc. for $C_{48}H_{54}N_7O_7P$ (871.98): C 66.12, H 6.24, N 11.24; found: C 66.47, H 6.31, N 11.18.

*(2′*S*)-2′-Deoxy-6-*O*-(2,6-dichlorophenyl)-5′-*O*-(4,4′-dimethoxytrityl)-2′-*C*-methylinosine* (**17**). To a soln. of **13** (65 mg, 0.16 mmol) in anh. pyridine (0.8 ml) and Et_3N (32.5 µl, 1.5 equiv.), 4,4′-dimethoxytrityl chloride (64.5 mg, 1.2 equiv.) and DMAQ (3 mg, 0.15 equiv.) were added with exclusion of moisture. The mixture was stirred at r.t. for 36 h, then quenched with H_2O, and evaporated. The residue was partitioned between CH_2Cl_2/sat. $NaHCO_3$ soln., the org. layer washed with H_2O, dried (Na_2SO_4), and evaporated, and the crude product purified by CC (CH_2Cl_2/MeOH/Et_3N 98:1:1): pure **17** (95 mg, 84%). ^{1}H-NMR ($CDCl_3$): 0.70 (*d*, *J*(2′,Me) = 7, Me–C(2′)); 2.62 (*m*, H–C(2′)); 3.62 (*m*, 2 H–C(5′)); 3.67 (*s*, 2 MeO); 3.92–4.25 (*m*, H–C(4′), H–C(3′), OH); 6.50 (*d*, *J*(1′,2′) = 7, H–C(1′)); 6.82 (*d*, *J* = 9, $(MeO)_2Tr$); 7.32–7.50 (*m*, 1 H, $(MeO)_2Tr$, 2 H_m, H_p); 8.21 (*s*, H–C(2)); 8.37 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 11.1 (*Me*–C(2′)); 45.3 (C(2′)); 55.2 ((*Me*O)$_2Tr$); 63.2 (C(5′)); 75.9 (C(3′)); 83.4 (C(4′)); 86.6[1]) (C(1′)); 86.9[2]) (*C*Ar$_3$ (MeO)$_2Tr$); 113.3 ((MeO)$_2Tr$); 121.1 (C(5)); 127.0, 127.1, 127.9, 128.2, 128.8, 129.4, 130.0, 135.6, 135.7 ((MeO)$_2Tr$, $C_6H_3Cl_2$); 142.3 (C(8)); 144.3 ((MeO)$_2Tr$); 145.4 ($C_6H_3Cl_2$); 151.7 (C(2)); 152.9 (C(4)); 158.3 (C(6)); 158.6 ((MeO)$_2Tr$). Anal. calc. for $C_{38}H_{34}Cl_2N_4O_6$ (713.62): C 63.96, H 4.80, N 7.85; found: C 64.22, H 4.84, N 7.81.

*(2′*S*)-2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-2′-*C*-methylinosine* (**18**). The soln. of **17** (90 mg, 0.13 mmol) in MeCN (2.0 ml) was treated with 2-nitrobenzaldehyde oxime (182 mg, 5.5 equiv.) and tetramethylguanidine (0.125 ml, 5 equiv.). The mixture was stirred under reflux for 18 h. As TLC still showed the presence of starting material, additional amounts of 2-nitrobenzaldehyde oxime (133 mg; 4 equiv.) and tetramethylguanidine (0.1 ml, 4 equiv.) were added, and the mixture was refluxed for another 24 h. Evaporation yielded a brown oil that was partitioned between H_2O and CH_2Cl_2. The org. layer was dried and evaporated and the crude product purified by CC (silica gel, CH_2Cl_2/MeOH 97:2 → 84:15 (1% Et_3N)): pure **18** (49.5 mg, 67%). ^{1}H-NMR ($CDCl_3$): 0.75 (*d*, *J*(2′,Me) = 7, Me–C(2′)); 2.82 (*m*, H–C(2′)); 3.52 (*m*, 2 H–C(5′)); 3.62 (br. *s*, OH); 3.81 (*s*, 2 MeO); 4.05 (*m*, H–C(4′), H–C(3′)); 6.41 (*d*, *J*(1′,2′) = 7, H–C(1′)); 6.83 (*d*, *J* = 9, 4 H, $(MeO)_2Tr$); 7.20–7.50 (*m*, 9 H, $(MeO)_2Tr$); 7.92 (*s*, H–C(2)); 8.09 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 10.8 (*Me*–C(2′)); 45.9 (C(2′)); 55.1 ((*Me*O)$_2$Tr); 63.0 (C(5′)); 74.6 (C(3′)); 83.6 (C(4′)); 86.2[2]) (C(1′)); 86.6[2]) (*C*Ar$_3$ (MeO)$_2Tr$)); 113.1

((MeO)$_2$*Tr*); 126.8 (C(5)); 126.8, 127.7, 128.1, 130.0, 135.6, 135.7 ((MeO)$_2$*Tr*); 138.7 (C(8)); 144.3 ((MeO)$_2$*Tr*); 144.8 (C(2)); 148.1 (C(4)); 158.3 (C(6)); 158.5 ((MeO)$_2$*Tr*). Anal. calc. for $C_{32}H_{32}N_4O_6$ (568.63): C 67.59, H 5.67, N 9.85; found: C 67.83, H 5.70, N 9.81.

*(2′-*S*)-2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-2′-*C*-methylinosine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**19**). Compound **18** (40 mg, 0.07 mmol) was dried by evaporation of MeCN. The foam was dissolved in dry 1,2-dichloroethane (0.4 ml) containing ^{i}Pr$_2$EtN (0.025 ml, 2 equiv.) under Ar, and the soln. was cooled in an ice-bath. Within 2 min, 2-cyanoethyl diisopropylphosphoramidochloridite (0.025 ml, 2 equiv.) was added dropwise, and the mixture was stirred at r.t. for 3 h. Then an excess of 2-cyanoethyl diisopropylphosphoramidochloridite (0.02 ml, 1.6 equiv.) was added and stirring continued for 2 h. The reaction was quenched by addition of CH_2Cl_2 and the soln. washed with 5% aq. $NaHCO_3$ soln. and brine. The org. layer was dried and evaporated and the crude product purified by CC ($CH_2Cl_2 \rightarrow CH_2Cl_2$/MeOH 95 :5 (1% Et_3N)): pure **19** (32 mg, 60%). ^{1}H-NMR ($CDCl_3$): 0.80 (*d*, *J*(2′,Me) = 7, Me–C(2′)); 1.21–1.32 (*m*, 2 *Me*$_2$CH); 2.60 (*m*, H–C(2′)); 2.97 (*m*, CH_2CH_2CN); 3.50 (*m*, CH_2CH_2CN, 2 H–C(5′)); 3.75 (*s*, 2 MeO); 4.02 (*m*, 2 Me_2CH), H–C(4′), H–C(3′)); 6.41 (*d*, *J*(1′,2′) = 7, H–C(1′)); 6.83 (*d*, *J* = 9, 4 H, (MeO)$_2$*Tr*); 7.20–7.52 (*m*, 9 H, (MeO)$_2$*Tr*); 7.90 (*s*, H–C(2)); 8.12 (*s*, H–C(8)). ^{13}C-NMR ($CDCl_3$): 11.1 (*Me*–C(2′)); 21.5 (CH_2CH_2CN); 24.3, 24.7, 25.4 (*Me*$_2$CH); 42.1, 43.2 (Me_2CH); 45.2 (C(2′)); 55.3 ((*Me*O)$_2$*Tr*); 56.7 (CH_2CH_2CN); 62.8 (C(5′)); 74.9 (C(3′)); 83.4 (C(4′)); 87.2[2]) (C(1′)); 86.8[2]) (*C*Ar$_3$ ((MeO)$_2$Tr)); 113.0 ((MeO)$_2$*Tr*); 117.0 (CN); 126.5 (C(5)); 126.9, 127.7, 128.0, 130.2, 135.4, 135.7 ((MeO)$_2$*Tr*); 138.4 (C(8)); 144.5 ((MeO)$_2$*Tr*); 144.6 (C(2)); 148.3 (C(4)); 158.1 (C(6)); 158.7 ((MeO)$_2$*Tr*). ^{31}P-NMR ($CDCl_3$): +147.5, 149.4. Anal. calc. for $C_{41}H_{49}N_6O_7P$ (768.85): C 64.05, H 6.42, N 7.29; found: C 64.32, H 6.51, N 7.23.

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