Protection of the guanine residue during synthesis of 2′-*O*-alkylguanosine derivatives

Morten Grøtli,[a] Mark Douglas,[a] Barbro Beijer,[b] Ramon Güimil García,[a] Ramon Eritja[a] and Brian Sproat*[,b]

[a] *EMBL, Meyerhofstrasse 1, D-69117 Heidelberg, Germany*
[b] *VPI GmbH, Olenhuser Landstrasse 20 a, D-37124 Rosdorf, Germany*

Highly selective 2′-*O*-alkylation of 3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine has been achieved by using an alkyl halide and a sterically hindered strong organic base, when the 6-O atom is protected with either a 2-nitrophenyl or a *tert*-butyldiphenylsilyl group prior to the alkylation. A minimum of chromatography is required, the yields are high and none of the unwanted isomer is produced. Moreover, the highly versatile intermediates enable the synthesis of several new 2′-*O*-alkylguanosine derivatives as well as base-modified analogues.

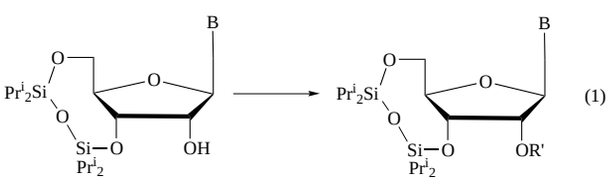
Introduction

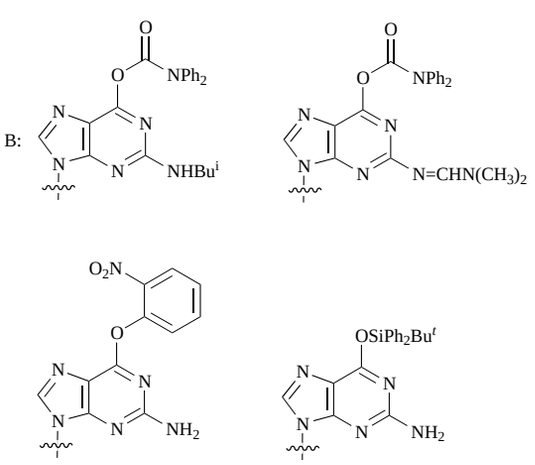
Oligonucleotide analogues are promising therapeutic agents for treatment of infectious diseases and cancers by acting as inhibitors of gene expression.[1,2] To be able to inhibit the gene expression the oligonucleotide must reach the interior of the cell unaltered. In order to do so they should be stable towards cellular nucleases and be able to penetrate through the cell membrane. In addition they have to hybridise with appropriate specificity and affinity to the complementary target nucleic acid. In order to meet all these requirements it is necessary for normal oligonucleotides to be chemically modified in a suitable manner.

2′-*O*-Alkyl ethers of oligoribonucleotides are oligonucleotide analogues that exhibit high resistance to both DNA- and RNA-specific nucleases and form hybrids of high thermal stability with complementary RNA.[3] These analogues have proven to be valuable compounds for antisense experiments and associated applications in biochemistry and molecular biology.[3–6] A considerable effort has therefore been directed towards developing efficient alkylation reactions that yield 2′-*O*-alkylribonucleoside building blocks. We recently reported an efficient route for the preparation of 2′-*O*-methyl- and 2′-*O*-allyl-guanosine derivatives.[7] In the present paper a detailed description of the synthesis of 2′-*O*-alkylguanosine derivatives is presented together with an alternative route that affords amide-protected derivatives. Moreover, several 2′-*O*-alkyl ethers of guanosine are described for the first time.

Results and discussion

A survey of the literature revealed a variety of problems encountered in the preparation and isolation of 2′-*O*-alkylguanosine derivatives. Early methods of methylation using diazomethane on unprotected or partially protected nucleosides[8] suffered from low yields and poor selectivity, giving mixtures of O- and N-methylation. The use of sodium hydride and an alkyl iodide on partially protected guanosine enhanced the selectivity for alkylation of the 2′-hydroxy function.[9] However, the separation of the 2′-*O*-methyl from the 3′-*O*-methyl ribonucleosides required subsequent protection of the exocyclic amino function and 5′-hydroxy moiety.[9] A major development was that of Hodge and Sinha[10] utilising an alkyl iodide and silver oxide as catalyst for mono-alkylation of the 2′,3′-diol moiety of cytidine. The 2′-*O*-alkylcytidine products could be converted into 2′-*O*-alkyluridines in one step. However, the silver oxide-catalysed alkylation procedure cannot be transferred to purine ribosides.[11] Specific alkylation of the 2′-hydroxy function has been reported using 3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl) (TIPDS)-protected ribonucleosides.[11–16] Other methods for the preparation of 2′-*O*-alkylguanosine derivatives utilised several additional protection and deprotection steps and resulted in low overall yields.[17]

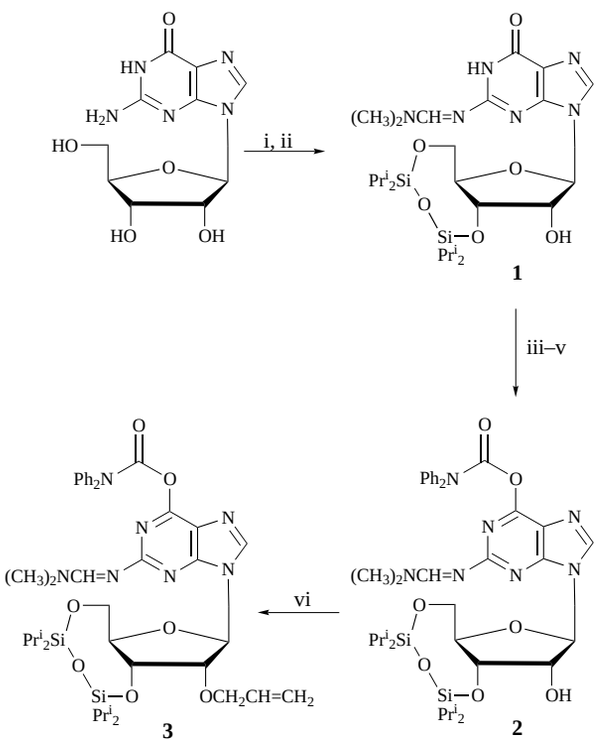




Different protecting groups tested in the present work, for protection of the guanine moiety during synthesis of 2′-*O*-alkylguanosine derivatives

The reaction schemes originally developed by Sproat *et al*. for preparing 2′-*O*-alkylguanosine phosphoramidite monomers are multistep and involve rather complex procedures for protection and deprotection.[11,14] We reckoned that it should be possible to protect the nucleobase in a much simpler way. Protection of the nucleobase is required, since alkylation of unprotected guanosine preferentially occurs at the base moiety.[9] Several different protecting groups were tested [equation (1)]. Initially, the diphenylcarbamoyl (DPC) group was selected as it also offers a guanosine 6-O-protection that is compatible with the oligonucleotide synthesis.[18,19] To ensure exclusive alkylation

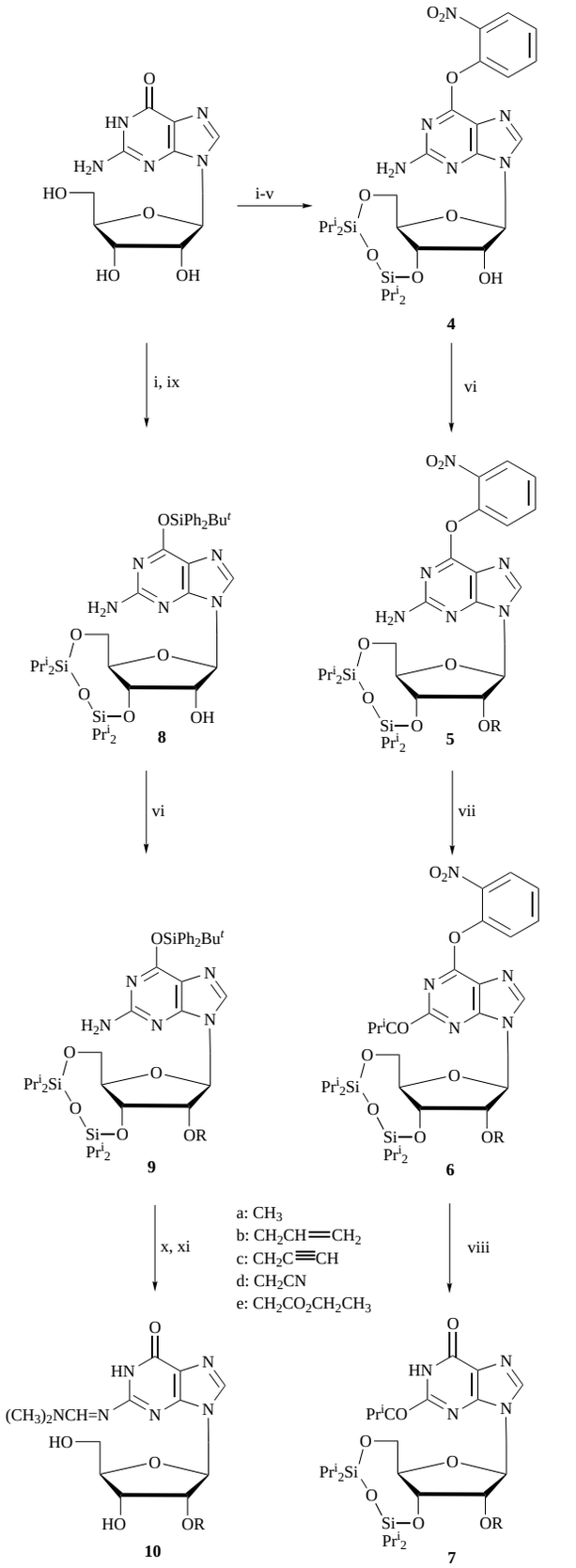
of the ribose 2′-hydroxy group, the Markiewicz protection protocol[20] for simultaneous blocking of the 5′- and 3′-hydroxy groups was used. Thus, guanosine was first treated with propionic anhydride, carbamoylated on the lactam 6-O and treated with aq. 2 mol dm^{-3} NaOH before it was protected with the Markiewicz disiloxane reagent, all according to the procedure of Kamimura *et al.*[18,19] Alkylation using previously described conditions,[11] with 2-*tert*-butylimino-2-diethylamino-1,3-dimethylperhydro-1,3,2-diazaphosphorine (BEMP) and methyl iodide, resulted in a mixture of 2-N- and 2′-O-methylation (data not shown). This result indicated that amide groups do not give the required protection of the exocyclic amino function under these alkylation conditions. For this reason an amidine-protection group was tested (Scheme 1). Thus, guanosine was



Scheme 1 *Reagents and conditions*: i, *N*,*N*-dimethylformamide dimethyl acetal in MeOH, room temp., 8 h; ii, TIPDSCl, imidazole in DMF, room temp., 1.5 h; iii, TMSCl, triethylamine in dichloromethane, room temp., 10 min; iv, DPCCl, DIPEA and DMAP in pyridine, room temp., 12 h; v, PTSA monohydrate in THF–dichloromethane, room temp., 2 min; vi, allyl ethyl carbonate, tris(dibenzylideneacetone)dipalladium(0), 1,4-bis(diphenylphosphino)butane in THF, reflux, 30 min

treated with an excess of *N*,*N*-dimethylformamide dimethyl acetal[21] in methanol. Subsequent Markiewicz protection afforded compound **1** in 86% yield. The DPC protecting group was introduced to give compound **2** in 71% yield. However, it was not possible to introduce the alkyl group on compound **2** by using the BEMP/alkyl halide system. No product formation was observed on TLC. On the other hand, allylation of compound **2** by the palladium-catalysed allylation procedure[13] gave an 86% isolated yield of compound **3**. This reaction proceeds through a rather bulky π-allyl-palladium complex but the amidine protection does not interfere as observed during the alkylation with BEMP.

Next, the possibility of achieving selective 2′-O-alkylation without any 2-N-protection was examined (Scheme 2). This approach excluded the use of the DPC protection group. Initial experiments showed that the introduction of the diphenylcarbamoyl protection on 3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine proceeded very sluggishly and resulted in low yields (data not shown). Reese and Stone have used the 2-nitrophenyl group successfully as a protecting group for the guanine residue during oligoribonucleotide synthesis.[22] We

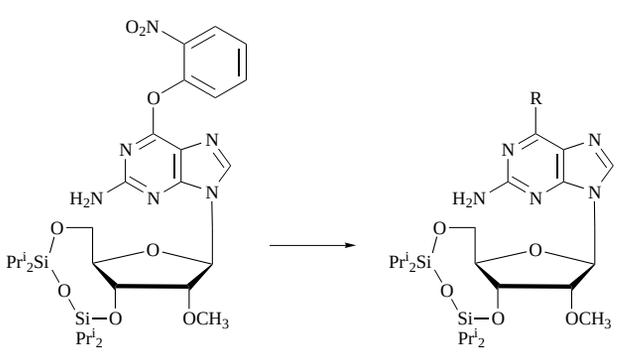


a: CH_3
b: $CH_2CH{=}CH_2$
c: $CH_2C{\equiv}CH$
d: CH_2CN
e: $CH_2CO_2CH_2CH_3$

Scheme 2 *Reagents and conditions*: i, TIPDSCl, imidazole in DMF, room temp., 1.5 h; ii, HMDS, 40 °C, 10 min; iii, mesitylene-2-sulfonyl chloride DMAP, triethylamine in dichloromethane, room temp., 15 h; iv, 2-nitrophenol, DABCO, triethylamine in acetonitrile, 80 °C, 30 min; v, PTSA monohydrate in THF–dichloromethane, room temp., 2 min; vi, BEMP, alkyl halide in MeCN; vii, isobutyryl chloride, DMAP in pyridine, 40 °C, 8 h; viii, *N*,*N*,*N*′,*N*′-tetramethylguanidine, 2-nitrobenzaldehyde oxime in acetonitrile, room temp., 16 h; ix, TBDPSCl, DMAP, triethylamine in dichloromethane, room temp., 8 h; x, TBAF in THF, room temp., 15 min; xi, *N*,*N*-dimethylformamide dimethyl acetal in MeOH, room temp., 12 h

therefore undertook the synthesis of 6-*O*-(2-nitrophenyl)-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine, compound **4**. The 2′-hydroxy group of 3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine was blocked as its trimethylsilyl (TMS) ether[23] and the product was allowed to react with mesitylenesulfonyl chloride in dichloromethane containing triethylamine and 4-(dimethylamino)pyridine (DMAP). The resulting 6-O-(mesitylene-2-sulfonyl) derivative was then heated at 80 °C with an excess of 2-nitrophenol and 1,4-diazabicyclo[2.2.2]octane (DABCO) in acetonitrile containing triethylamine, followed by cleavage of the TMS ether, to give compound **4** in 75% overall yield. Alkylation of compound **4** with 2 mol equiv. of propargyl bromide and 2 mol equiv. of BEMP gave compound **5c** in 71% isolated yield. No 2-N-alkylation was observed and ring opening of the disiloxane bridge was minimal. The 2′-*O*-methyl, 2′-*O*-cyanomethyl and 2′-*O*-(ethoxycarbonyl)methyl analogues **5a**, **5d** and **5e** were obtained, in a similar fashion, in 70 and 63% yield respectively. The cyanomethyl group is particularly interesting since it can be reduced to aminoethyl or hydrolysed to the amide or carboxylate.[24] Moreover the ester moiety of 2′-*O*-(ethoxycarbonyl)methylribonucleotide monomers can be converted into a variety of useful functionalities by performing an appropriate nucleophilic displacement reaction upon it.[25] The 2′-*O*-(ethoxycarbonyl)methyl group can also be reduced to the alcohol, enabling the synthesis of 2′-*O*-(2-sulfanylethyl)ribonucleotides which can be readily tagged with a variety of useful reporter groups.[26] We also confirmed that the palladium-catalysed allylation of compound **4** with 1 mol equiv. of allyl ethyl carbonate led to compound **5b** in 88% isolated yield. An excess of allyl ethyl carbonate led to a diallylated product (data not shown). Treatment of compounds **5a–5e** with isobutyryl chloride–DMAP in pyridine at 40 °C afforded **6a–6e** in good yields (76–86%). When these compounds were treated with 3 mol equiv. of *N*,*N*,*N*′,*N*′-tetramethylguanidine and 3 mol equiv. of 2-nitrobenzaldehyde oxime in acetonitrile at room temperature overnight, compounds **7a–7e** were obtained in high yields. The overall yield for the 8-step synthesis was between 29 and 42% based on guanosine.

Compounds **5a** and **5b** may well be useful intermediates for the preparation of 2′-*O*-alkylguanosine analogues modified at position 6 (Scheme 3). In fact, compound **5a** was readily con-



Scheme 3 *Reagents and conditions*: **11**, H_2NNH_2 in THF, room temp., 48 h; **12**, as **11**, then Ag_2O in THF–water, reflux, 2 h; **13**, NH_3 in THF, 70 °C, 5 days

verted into 2-amino-6-hydrazino-2′-*O*-methyl-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)purine riboside **11** in 72% isolated yield (data not shown).[27] Subsequent oxidative elimination of the hydrazino group with Ag_2O gave the 2-aminopurine derivative **12** in 68% isolated yield.[28] Displacement of 2-nitrophenoxide from compound **5a** by ammonia–tetrahydrofuran (THF) in a stainless steel bomb for 5 days at 70 °C gave the 2′-*O*-methyl-3′,5′-*O*-(tetraisopropylidisiloxane-1,3-diyl)riboside derivative **13** in 99% yield.[11]

The DMAP–triethylamine–dichloromethane system used above to synthesize the 6-*O*-(mesitylene-2-sulfonyl) derivative has also been applied to the 6-O-silylation of guanosine derivatives.[29] The combination of Markiewicz protection for simultaneous blocking of the 5′- and 3′-hydroxy groups and 6-O-silyl protection seemed very attractive since both groups can be removed simultaneously under mild conditions after the alkylation. Thus, the 6-O-silylation of 3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine using *tert*-butyl(chloro)diphenylsilane (TBDPSCl)[30] was examined (Scheme 2). 3′,5′-*O*-(Tetraisopropyldisiloxane-1,3-diyl)guanosine was converted into the highly versatile 6-O-TBDPS derivative **8**, using TBDPSCl, DMAP and triethylamine in dichloromethane at room temperature. The reaction gave selective 6-O-silylation without any observable 2′-O-silylation. The 6-O-silylated product was found to be formed essentially quantitatively by monitoring the reaction on TLC. Other reagents, like *tert*-butoxy(chloro)diphenylsilane and *tert*-butyl(chloro)dimethylsilane, also yielded 6-O-silylated products quantitatively. However, none of the compounds was stable to chromatography on silica gel; only compound **8** partly survived silica gel chromatography (47% yield). The other two 6-O-silylated derivatives were completely unstable to silica gel chromatography and were excluded from further use due to their low stability. However, since the synthesis of the 6-O-TBDPS derivative **8** was essentially quantitative it was used further without any purification. It is worth noting that the IR spectrum lacked the usual carbonyl band at 1700 cm^{-1} thereby indicating that the lactam function was in the enol form and that the TBDPS group was attached to the 6-O atom.[7]

Silylation with TBDPSCl is normally performed in the presence of a base which acts as an acceptor for HCl but which also modifies the reactivity of the silylating agent. Further, the choice of an appropriate solvent and catalyst enhances the silylation potential of the reaction mixture. We therefore examined the silylation with TBDPSCl under different conditions. By using triethylamine and DMAP as a catalyst in dichloromethane good yields were obtained. When the solvent was changed from dichloromethane to pyridine no product formation was observed on TLC. This was in part due to the problem of dissolving 3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)-guanosine in pyridine. TBDPSCl–DMAP–triethylamine with DMF as a solvent gave a similar result with no formation of either the 2′-O-silylated or the 6-O-silylated derivative. When imidazole was added to the TBDPSCl–DMAP–triethylamine–dichloromethane system the selectivity changed almost completely from 6-O-silylation to 2′-O-silylation. It is likely that the imidazole is assisting the removal of the 2′-OH proton as described for the intermolecular transesterification of ethyl 2-(hydroxymethyl)benzoates.[31] It is therefore essential to remove all the imidazole from the introduction of the TIPDS group. Addition of imidazole to the TBDPSCl–DMAP–triethylamine–pyridine system resulted in formation of the 2′-O-silylated derivative in 33% isolated yield after 8 h (data not shown). By replacing pyridine with DMF the reaction proceeded faster and gave the 2′-O-silylated derivative in 71% isolated yield (data not shown). However, neither of the two reaction mixtures produced any observable amount of the 6-O-silylated derivative.

It was found that highly selective 2′-O-alkylation of compound **8** could be achieved by using 5 mol equiv. of alkyl halide and 2 mol equiv. of the sterically hindered strong organic base BEMP. No alkylation on the base moiety was observed. Ring opening of the disiloxane bridge was minimal and compounds **9a**–**9e** were formed (no data given). Subsequent desilylation with tetrabutylammonium fluoride (TBAF) removed the disiloxane bridge and the TBDPS group. In the case of the 2′-*O*-[(ethoxycarbonyl)methyl]guanosine derivative a 1:1 mixture of TBAF and acetic acid in THF was used to prevent cleavage of the ester function. In order to avoid the transient protection

procedure[24] we decided to block the exocyclic amino group of the 2′-*O*-alkylguanosine derivatives by use of an amidine moiety. Thus the 2′-*O*-alkyl ethers of guanosine were treated with dimethylformamide dimethyl acetal[21] to give compounds **10a**–**10e**. The overall yield for the 5-step synthesis was between 47 and 51% based on guanosine.

In summary we have demonstrated that highly selective 2′-O-alkylation of 3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine can be achieved by using an alkyl halide and BEMP, when the 6-O atom is protected with either a 2-nitrophenyl or a TBDPS group prior to the alkylation. Although the methodology using 2-nitrophenyl as a protecting group is multistep, the overall yields are higher than in previously published work and it has the flexibility of enabling the introduction of any desired protecting group on the exocyclic amino function, without adding additional steps to the synthesis. Moreover, the methodology gives useful intermediates for the preparation of 2′-*O*-alkylguanosine analogues modified at 6 position. By using TBDPS for 6-O protection, 2′-*O*-alkyl-2-*N*-(dimethylaminomethylene)guanosines can be obtained in only 5 steps starting from guanosine. A minimum of chromatography is required, the yields are high and none of the unwanted isomer is produced.

Experimental

NMR spectra were recorded on a Bruker AM 250 spectrometer at the following frequencies: 250 MHz for ^{1}H NMR and 62 MHz for ^{13}C NMR. Chemical shifts (δ) are reported in parts per million (ppm) downfield relative to the internal tetramethylsilane standard, and *J*-values are in Hz. Mass spectra were recorded using either Fast Atom Bombardment (FAB) or Electrospray (ES) ionisation, UV spectra were obtained on a Varian Cary 3 spectrophotometer. The silica gel (35–70 μm) used for column chromatography was purchased from SDS. TLC was carried out on Merck DC Kieselgel 60 F-254 aluminium sheets. 1,3-Dichloro-1,1,3,3-tetraisopropyldisiloxane (TIPDSCl) was obtained from Ifotam (Lodz, Poland). All reagents used were of the highest available purity. Anhydrous solvents were purchased from SDS.

2-*N*-(Dimethylaminomethylene)-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 1

Guanosine (3.66 g, 14 mmol) was suspended in dry methanol (100 cm^3) and dimethylformamide dimethyl acetal (10 cm^3, 76.0 mmol) was added. The mixture was stirred overnight at room temperature and the desired product was removed by filtration. The solid was washed with methanol and dried over phosphorus pentaoxide *in vacuo*. This product was treated with imidazole (5.3 g, 77.85 mmol) and TIPDSCl (4.9 cm^3 15.54 mmol) as a solution in dry dimethylformamide (DMF) (100 cm^3). The reaction mixture was left at room temperature. TLC showed complete reaction after 1.5 h. The solvent was evaporated off under reduced pressure. The residue was dissolved in ethyl acetate (250 cm^3), washed with 1 mol dm^{-3} aq. sodium hydrogen carbonate (250 cm^3), the organic layer was separated, dried (Na_2SO_4), filtered, and evaporated *in vacuo*. Chromatography of the crude product on silica gel (170 g) and elution with a gradient of ethanol 0–5% in dichloromethane afforded pure compound **1** as a solid foam (7.72 g, 86.2%), R_f 0.37 on TLC in ethanol–dichloromethane (5:95 v/v). The spectroscopic data were in agreement with previously published data.[32]

2-*N*-(Dimethylaminomethylene)-6-*O*-diphenylcarbamoyl-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 2

Compound **1** (3.0 g, 5.16 mmol) was dissolved in dry dichloromethane (100 cm^3) and triethylamine (3 cm^3, 21.54 mmol) and TMSCl (2.0 cm^3, 15.81 mmol) were added to the stirred solution with exclusion of moisture. TLC showed complete reaction after 10 min at room temperature. The reaction mixture was poured into vigorously stirred 1.1 mol dm^{-3} aq. sodium hydrogen carbonate (200 cm^3), and then the separated organic phase was dried (Na_2SO_4), filtered, and evaporated *in vacuo*. The foam was dried by evaporation of dry pyridine (25 cm^3) and dissolved in dry pyridine (75 cm^3) under argon. Diisopropylethylamine (DIPEA) (4 cm^3, 22.29 mmol), DMAP (0.2 g, 1.64 mmol) and diphenylcarbamoyl chloride (DPCCl) (2.4 g, 10.36 mmol) were added to the stirred mixture with exclusion of moisture. The reaction mixture was left at room temperature for 12 h. TLC then showed complete reaction. The solvent was evaporated off under reduced pressure. The residue was dissolved in ethyl acetate (250 cm^3), the solution was washed with 1 mol dm^{-3} aq. sodium hydrogen carbonate (250 cm^3), and the organic layer was separated, dried (Na_2SO_4), filtered, and evaporated *in vacuo*. The residual was dissolved in dichloromethane (150 cm^3). A solution of toluene-*p*-sulfonic acid (PTSA) monohydrate (2.0 g, 10.52 mmol) in THF (10 cm^3) was added with stirring of the mixture, and after 2 min the acid was quenched by addition of triethylamine (2 cm^3, 14.36 mmol). The reaction mixture was poured into vigorously stirred 1 mol dm^{-3} aq. sodium hydrogen carbonate (200 cm^3), and then the separated organic phase was dried (Na_2SO_4), filtered, and the solvent was removed *in vacuo*. Chromatography of the crude product on silica gel (80 g) and elution with ethyl acetate–hexane (1:1 and 2:1 v/v) afforded *pure compound* **2** as a solid foam (2.85 g, 71.2%), R_f 0.44 on TLC in ethanol–dichloromethane (5:95 v/v) (Found: C, 58.87; H, 6.95; N, 12.74. $C_{38}H_{53}N_7O_7Si_2$ requires C, 58.81; H, 6.90; N, 12.64%); ν_{max}(KBr)/cm^{-1} 2947 and 2869 (CH st), 1715 (carbamoyl C=O), 1628 and 1578 (C=C, N=C); δ_H($CDCl_3$) 8.42 (1 H, s, amidine CH), 7.93 (1 H, s, H-8), 7.10–7.40 (10 H, Ph), 6.05 (1 H, d, *J* 5.4, H-1′), 4.58 (1 H, m, H-3′), 4.36 (1 H, d, *J* 5.1, H-2′), 4.11 (3 H, m, H-4′ and H_2-5′), 3.16 (3 H, s, amidine CH_3), 3.06 (3 H, s, amidine CH_3) and 1.07 (28 H, m, Pr^i); δ_C($CDCl_3$) 162.55 (carbamoyl C=O), 158.68 (amidine CH), 155.54 (C-6), 154.91 (C-2), 150.77 (C-4), 141.99 (Ph C-1), 140.46 (C-8), 128.84 and 126.74 (Ph C-2, -3, -5 and -6), 126.45 (Ph C-4), 120.72 (C-5), 88.40 (C-1′), 81.89 (C-4′), 74.98 (C-2′), 70.30 (C-3′), 61.33 (C-5′), 40.83 and 34.89 (amidine CH_3), 17.23–16.71 [$CH(CH_3)_2$] and 13.17, 12.86, 12.77 and 12.40 [$CH(CH_3)_2$]; ES-MS *m/z* 776.2 (M^+).

2′-*O*-Allyl-2-*N*-(dimethylaminomethylene)-6-*O*-diphenylcarbamoyl-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 3

Tris(dibenzylideneacetone)dipalladium(0) (6 mg, 0.007 mmol) and 1,4-bis(diphenylphosphino)butane (21 mg, 0.05 mmol) were suspended in dry THF (2 cm^3) under argon. A solution of compound **2** (0.59 g, 0.76 mmol) and allyl ethyl carbonate (0.2 cm^3, 1.54 mmol) in dry THF (18 cm^3) was added, and the mixture was refluxed under argon for 30 min. TLC showed complete reaction and solvent was removed *in vacuo*. The crude product was purified by column chromatography on silica gel (20 g) and elution with a gradient of ethyl acetate from 33 to 50% in hexane. The *pure product* **3** was obtained as a foam (0.53 g, 85.5%) of R_f 0.10 on TLC in hexane–ethyl acetate (1:1 v/v) (Found: C, 60.45; H, 7.07; N, 12.06. $C_{41}H_{57}N_7O_7Si_2$ requires C, 60.33; H, 7.05; N, 12.02%); ν_{max}(KBr)/cm^{-1} 3090 (allyl C=C), 2948 and 2868 (CH st), 1717 (carbamoyl C=O), 1629 and 1577 (C=C, N=C); δ_H($CDCl_3$) 8.44 (1 H, s, amidine CH), 7.90 (1 H, s, H-8), 7.11–7.43 (10 H, Ph), 6.10 (1 H, m, allyl CH), 6.03 (1 H, d, *J* 1.4, H-1′), 5.21 (2 H, m, allyl=CH_2), 4.59 (1 H, m, H-3′), 4.35 (1 H, d, *J* 5.1, H-2′), 4.07 (3 H, m, H-4′ and H_2-5′), 3.15 (3 H, s, amidine CH_3), 3.07 (3 H, s, amidine CH_3) and 1.08 (28 H, m, Pr^i); δ_C($CDCl_3$) 162.50 (carbamoyl C=O), 158.53 (amidine CH), 155.51 (C-6), 154.70 (C-2), 150.81 (C-4), 142.02 (Ph C-1), 140.24 (C-8), 134.22 (allyl CH), 128.79 and 126.73 (Ph C-2, -3, -5 and -6), 126.40 (Ph C-4), 120.90 (C-5), 117.22 (allyl =CH_2), 87.89 (C-1′), 81.24 (C-2′ and -4′), 71.68 (allyl CH_2), 71.69 (C-3′), 59.41 (C-5′), 40.78 and 34.92 (amidine CH_3), 17.15–16.73 [$CH(CH_3)_2$] and 13.25, 12.78 and 12.49 [$CH(CH_3)_2$]; ES-MS *m/z* 816.3 (M^+).

6-*O*-(2-Nitrophenyl)-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 4

Guanosine (7.58 g, 26.76 mmol) was dried by evaporation of dry pyridine (40 cm^3) and dissolved in dry DMF (100 cm^3) under argon. Imidazole (7.3 g, 107.20 mmol) and TIPDSCl tetraisopropyldisiloxane (9.5 cm^3, 30.12 mmol) were added to the stirred mixture with exclusion of moisture. The reaction mixture was left at room temperature. TLC showed complete reaction after 1.5 h. Hexamethyldisilazane (HMDS) was added dropwise and after 10 min the reaction mixture was concentrated *in vacuo* at 40 °C. The residue was dried under reduced pressure, dissolved in dichloromethane (400 cm^3), the solution was washed with 1 mol dm^{-3} aq. sodium hydrogen carbonate (400 cm^3), and the organic layer was separated, dried (Na_2SO_4), filtered, and evaporated *in vacuo*. The residue was dissolved in dry dichloromethane (150 cm^3), and triethylamine (18 cm^3, 129.25 mmol), DMAP (0.8 g, 6.55 mmol) and mesitylene-2-sulfonyl chloride (6.44 g, 29.45 mmol) were added to the stirred mixture with exclusion of moisture. TLC showed complete reaction after 15 h. The solvent was evaporated off under reduced pressure. The residue was dissolved in dichloromethane (400 cm^3), the solution was washed with 1 mol dm^{-3} aq. sodium hydrogen carbonate (400 cm^3), and the organic layer was separated, dried (Na_2SO_4), filtered, and evaporated *in vacuo*. 2-Nitrophenol (7.5 g, 53.91 mmol), DABCO (0.6 g, 5.35 mmol) and triethylamine (18 cm^3, 129.25 mmol) were dissolved in dry acetonitrile (50 cm^3) and added with exclusion of moisture to a stirred solution of the above residue in acetonitrile (100 cm^3). The reaction mixture was heated at 80 °C for 30 min. TLC showed complete reaction. The solvent was evaporated off *in vacuo*, the residual oil was treated with a solution of PTSA monohydrate (11.5 g, 60.46 mmol) in THF–dichloromethane (200 cm^3; 1:5 v/v) for 2 min at room temperature and the acid was then quenched by addition of triethylamine (10 cm^3, 71.80 mmol). The solution was diluted with dichloromethane (200 cm^3) and then poured into vigorously stirred 1 mol dm^{-3} aq. sodium hydrogen carbonate (400 cm^3). The organic layer was separated, dried (Na_2SO_4), filtered and evaporated *in vacuo*. The crude product was purified by column chromatography on silica gel (400 g) and elution with a gradient of 33 to 60% ethyl acetate in hexane. The *pure product* **4** was obtained as a yellow foam (12.89 g, 74.5% from guanosine), R_f 0.29 on TLC in hexane–ethyl acetate (1:1 v/v) (Found: C, 52.13; H, 6.59; N, 13.07. $C_{28}H_{42}N_6O_8Si_2$ requires C, 51.98; H, 6.56; N, 12.99%); ν_{max}(KBr)/cm^{-1} 3347br (NH, OH), 2947 and 2869 (CH st), 1629 and 1576 (C=C, N=C) and 1533 (NO_2); δ_H($CDCl_3$) 10.03 (2 H, br s, NH_2), 8.11 (1 H, s, H-8), 7.24–7.96 (4 H, ArH), 6.03 (1 H, d, *J* 5.4, H-1′), 4.59–4.19 (5 H, m, H-2′, -3′, -4′ and H_2-5′) and 1.09 (28 H, m, Pr^i); δ_C($CDCl_3$) 158.90 (C-2 and -6), 154.11 (C-4), 145.33 (Ph, C-1), 142.41 (Ph, C-2), 138.55 (C-8), 134.39 (Ph, C-5), 125.97 (Ph, C-3), 125.31 (Ph, C-4 and -6), 115.54 (C-5), 88.67 (C-1′), 81.52 (C-4′), 74.82 (C-2′), 70.07 (C-3′), 61.01 (C-5′), 17.14 and 16.80 [$CH(CH_3)_2$] and 13.18, 12.80 and 12.40 [$CH(CH_3)_2$]; ES-MS *m/z* 647.3 (M^+).

2′-*O*-Methyl-6-*O*-(2-nitrophenyl)-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 5a

Compound **4** (1.50 g, 2.32 mmol) was dried by evaporation of dry acetonitrile (10 cm^3) and was dissolved in anhydrous acetonitrile (30 cm^3) under argon. BEMP (1.4 cm^3, 4.84 mmol) followed immediately by methyl iodide (0.43 cm^3, 4.64 mmol) were added to the stirred mixture with exclusion of moisture. TLC showed more or less complete reaction after 3 h. The reaction was quenched with methanol (0.7 cm^3) and the solvent was evaporated off *in vacuo*. The residue was dissolved in dichloromethane (100 cm^3), the solution was washed with 0.5 mol dm^{-3} aq. sodium phosphate buffer, pH 7 (100 cm^3), and the organic layer was separated, dried (Na_2SO_4), filtered, and evaporated *in vacuo*. The crude product was purified by column chromatography on silica gel (20 g) and elution with hexane–ethyl acetate (2:1 v/v). The *pure product* **5a** was obtained as a foam (1.01 g, 61.2%) of R_f 0.11 on TLC in hexane–ethyl acetate (2:1 v/v) (Found: C, 52.80; H, 6.76; N, 12.77. $C_{29}H_{44}N_6O_8Si_2$ requires C, 52.70; H, 6.72; N, 12.72%); ν_{max}(KBr)/cm^{-1} 3380 (NH), 2948 and 2868 (CH st), 1629 and 1578 (C=C, N=C) and 1534 (NO_2); δ_H($CDCl_3$) 10.10 (2 H, br s, NH_2), 8.13 (1 H, s, H-8), 7.96–7.24 (4 H, ArH), 6.03 (1 H, d, *J* 5.4, H-1′), 4.61–4.19 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 3.41 (3 H, s, OCH_3) and 1.11 (28 H, m, Pr^i); δ_C($CDCl_3$) 158.78 (C-2 and -6), 154.37 (C-4), 145.31 (Ph, C-1), 142.59 (Ph, C-2), 138.64 (C-8), 134.43 (Ph, C-5), 125.87 (Ph, C-3), 125.41 (Ph, C-4 and -6), 115.65 (C-5), 87.71 (C-1′), 81.87 (C-4′), 80.57 (C-2′), 69.81 (C-3′), 59.70 (C-5′), 58.03 (OCH_3), 17.33 [$CH(CH_3)_2$] and 13.21, 12.77 and 12.41 [$CH(CH_3)_2$]; FAB-MS *m/z* 662.1 $[M + H]^+$.

2′-*O*-Allyl-6-*O*-(2-nitrophenyl)-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 5b

Tris(dibenzylideneacetone)dipalladium(0) (6 mg, 0.007 mmol) and 1,4-bis(diphenylphosphino)butane (21 mg, 0.05 mmol) were suspended in dry THF (2 cm^3) under argon. A solution of compound **4** (0.75 g, 1.16 mmol) and allyl ethyl carbonate (0.17 cm^3, 1.31 mmol) in dry THF (18 cm^3) was added, and the mixture was refluxed under argon for 30 min. TLC showed complete reaction and the solvent was removed *in vacuo*. The crude product was purified by column chromatography on silica gel (20 g) and elution with a gradient of ethyl acetate from 20 to 33% in hexane. The *pure product* **5b** was obtained as a foam (0.70 g, 87.8%) of R_f 0.27 on TLC in hexane–ethyl acetate (2:1 v/v) (Found: C, 54.31; H, 6.78; N, 12.28. $C_{31}H_{46}N_6O_8Si_2$ requires C, 54.20; H, 6.76; N, 12.24%); ν_{max}(KBr)/cm^{-1} 3381 (NH), 2946 and 2869 (CH st), 1630 and 1577 (C=C, N=C) and 1532 (NO_2); δ_H($CDCl_3$) 10.05 (2 H, br s, NH_2), 8.11 (1 H, s, H-8), 7.96–7.27 (4 H, ArH), 6.12 (1 H, m, allyl CH), 6.05 (1 H, d, *J* 5.5, H-1′), 5.19 (2 H, m, allyl $=CH_2$), 4.61–4.20 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 3.78 (2 H, m, allyl CH_2) and 1.07 (28 H, m, Pr^i); δ_C($CDCl_3$) 158.73 (C-2 and -6), 154.16 (C-4), 145.46 (Ph, C-1), 142.53 (Ph, C-2), 138.44 (C-8), 134.49 (Ph, C-5), 134.19 (allyl CH), 125.97 (Ph, C-3), 125.35 (Ph, C-4 and -6), 117.31 (allyl $=CH_2$), 115.63 (C-5), 88.14 (C-1′), 81.21 (C-4′), 80.98 (C-2′), 71.66 (allyl OCH_2), 69.52 (C-3′), 59.81 (C-5′), 17.20 and 16.95 [$CH(CH_3)_2$] and 13.33, 12.85 and 12.51 [$CH(CH_3)_2$]; FAB-MS *m/z* 688.1 $[M + H]^+$.

6-*O*-(2-Nitrophenyl)-2′-*O*-propargyl-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 5c

Compound **4** (1.32 g, 2.04 mmol) was treated with BEMP (1.2 cm^3, 4.15 mmol) and propargyl bromide (0.37 cm^3, 4.15 mmol) according to the procedure for compound **5a**. The crude product was purified by column chromatography on silica gel (20 g) and elution with hexane–ethyl acetate (2:1 v/v). The *pure product* **5c** was obtained as a foam (0.99 g, 70.7%) of R_f 0.17 on TLC in hexane–ethyl acetate (2:1 v/v) (Found: C, 54.33; H, 6.55; N, 12.26. $C_{31}H_{44}N_6O_8Si_2$ requires C, 54.40; H, 6.49; N, 12.27%); ν_{max}(KBr)/cm^{-1} 3370 (NH), 3335 (propargyl C≡C), 2945 and 2868 (CH st), 1630 and 1576 (C=C, N=C) and 1534 (NO_2); δ_H($CDCl_3$) 10.02 (2 H, br s, NH_2), 8.13 (1 H, s, H-8), 7.96–7.24 (4 H, ArH), 6.03 (1 H, d, *J* 5.4, H-1′), 4.73 (2 H, AB, *J* 16, propargyl CH_2), 4.61–4.19 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 2.57 (1 H, s, propargyl CH) and 1.11 (28 H, m, Pr^i); δ_C($CDCl_3$) 158.77 (C-2 and -6), 154.21 (C-4), 145.46 (Ph, C-1), 142.56 (Ph, C-2), 138.49 (C-8), 134.46 (Ph, C-5), 126.01 (Ph, C-3), 125.40 (Ph, C-4, -6), 115.67 (C-5), 88.00 (C-1′), 81.14 (C-4′), 79.95 (C-2′), 79.15 (propargyl q C), 75.27 (propargyl CH), 69.66 (C-3′), 59.73 (C-5′), 58.21 (propargyl CH_2), 17.21 [$CH(CH_3)_2$] and 13.27, 12.85 and 12.44 [$CH(CH_3)_2$]; FAB-MS *m/z* 690.1 $[M + H]^+$.

2′-*O*-Cyanomethyl-6-*O*-(2-nitrophenyl)-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 5d

Compound **4** (1.50 g, 2.32 mmol) was treated with BEMP (1.4

cm^3, 4.84 mmol) followed by bromoacetonitrile (0.40 cm^3, 5.74 mmol) according to the procedure for compound **5a**. The crude product was purified by column chromatography on silica gel (20 g) and elution with hexane–ethyl acetate (2:1 and 3:2 v/v). The *pure product* **5d** was obtained as a foam (1.11 g, 69.8%) of R_f 0.16 on TLC in hexane–ethyl acetate (2:1 v/v) (Found: C, 52.61; H, 6.37; N, 14.33. $C_{30}H_{43}N_7O_8Si_2$ requires C, 52.53; H, 6.33; N, 14.30%); ν_{max}(KBr)/cm^{-1} 3376 (NH), 2946 and 2871 (CH st), 2180 (cyanomethyl CN), 1630 and 1577 (C=C, N=C) and 1531 (NO_2); δ_H($CDCl_3$) 10.05 (2 H, br s, NH_2), 8.15 (1 H, s, H-8), 7.96–7.24 (4 H, ArH), 6.03 (1 H, d, *J* 5.4, H-1′), 4.61–4.19 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 4.25 (2 H, *J* 15.5, CH_2CN) and 1.11 (28 H, m, Pr^i); δ_C($CDCl_3$) 158.98 (C-2 and -6), 153.88 (C-4), 145.31 (Ph, C-1), 142.44 (Ph, C-2), 138.00 (C-8), 134.56 (Ph, C-5), 126.16 (Ph, C-3), 125.43 (Ph, C-4 and -6), 115.74 (CH_2CN), 115.45 (C-5), 87.76 (C-1′), 81.73 (C-4′), 81.07 (C-2′), 69.52 (C-3′), 59.38 (C-5′), 56.15 (CH_2CN), 17.08 [$CH(CH_3)_2$] and 13.30, 12.75 and 12.46 [$CH(CH_3)_2$]; ES-MS *m/z* 686.1 (M^+).

2′-*O*-(Ethoxycarbonyl)methyl-6-*O*-(2-nitrophenyl)-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 5e

Compound **4** (1.50 g, 2.32 mmol) was treated with BEMP (1.4 cm^3, 4.84 mmol) and ethyl bromoacetate (0.65 cm^3, 5.86 mmol), according to the procedure for compound **5a**. The crude product was purified by column chromatography on silica gel (20 g) and elution with hexane–ethyl acetate (2:1 v/v). The *pure product* **5e** was obtained as a foam (1.07 g, 62.9%) of R_f 0.14 on TLC in hexane–ethyl acetate (2:1 v/v) (Found: C, 52.34; H, 6.62; N, 11.48. $C_{32}H_{48}N_6O_{10}Si_2$ requires C, 52.29; H, 6.60; N, 11.44%); ν_{max}(KBr)/cm^{-1} 3373 (NH), 2947 and 2868 (CH st), 1731 ([ethoxycarbonyl]methyl C=O), 1631 and 1578 (C=C, N=C) and 1532 (NO_2); δ_H($CDCl_3$) 10.12 (2 H, br s, NH_2), 8.12 (1 H, s, H-8), 7.98–7.23 (4 H, ArH), 6.02 (1 H, d, *J* 5.4, H-1′), 4.63–4.22 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 4.21 (2 H, AB, *J* 16, [ethoxycarbonyl]methyl CH_2), 3.42 (2 H, m, [ethoxycarbonyl]methyl CH_2), 1.78 (3 H, m, [ethoxycarbonyl]methyl CH_3) and 1.10 (28 H, m, Pr^i); δ_C($CDCl_3$) 169.61 ([ethoxycarbonyl]methyl C=O), 158.72 (C-2 and -6), 154.10 (C-4), 145.38 (Ph, C-1), 142.48 (Ph, C-2), 138.53 (C-8), 134.38 (Ph, C-5), 125.94 (Ph, C-3), 125.27 (Ph, C-4, -6), 115.59 (C-5), 87.86 (C-1′), 82.07 (C-4′), 80.68 (C-2′), 69.69 (C-3′), 68.10 ([ethoxycarbonyl]methyl CH_2), 60.61 ([ethoxycarbonyl]methyl CH_2), 59.71 (C-5′), 17.21 [$CH(CH_3)_2$], 13.27, 12.85 and 12.44 [$CH(CH_3)_2$] and 12.80 ([ethoxycarbonyl]-methyl CH_3); ES-MS *m/z* 733.1 (M^+).

2-*N*-Isobutyryl-2′-*O*-methyl-6-*O*-(2-nitrophenyl)-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 6a

Compound **5a** (1.00 g, 1.36 mmol) was dried by evaporation of dry pyridine (10 cm^3) and dissolved in anhydrous pyridine (20 cm^3) under argon. DMAP (0.020 g, 0.16 mmol) followed by isobutyryl chloride (0.25 cm^3, 2.41 mmol) were added to the stirred mixture with exclusion of moisture. The reaction was heated at 40 °C for 8 h. TLC showed more or less complete reaction. The reaction was quenched with methanol (0.5 cm^3) and solvent was evaporated off *in vacuo*. The residue was dissolved in dichloromethane (100 cm^3), the solution was washed with 1 mol dm^{-3} aq. sodium hydrogen carbonate (100 cm^3), and the organic layer was separated, dried (Na_2SO_4), filtered, and evaporated *in vacuo*. The crude product was purified by column chromatography on silica gel (25 g) and elution with hexane–ethyl acetate (4:1 and 3:1 v/v). The *pure product* **6a** was obtained as a foam (0.88 g, 80.6%) of R_f 0.14 on TLC in hexane–ethyl acetate (3:1 v/v) (Found: C, 52.60; H, 6.94; N, 11.57. $C_{33}H_{50}N_6O_9Si_2$ requires C, 52.57; H, 6.91; N, 11.50%); ν_{max}(KBr)/cm^{-1} 2948 and 2868 (CH st), 1630 and 1577 (C=C, N=C) and 1532 (NO_2); δ_H($CDCl_3$) 10.53 (1 H, br s, NH), 8.13 (1 H, s, H-8), 7.96–7.24 (4 H, ArH), 6.03 (1 H, d, *J* 5.4, H-1′), 4.61–4.19 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 3.41 (3 H, s, OCH_3), 2.15 [1 H, m, $(CH_3)_2CHCO$] and 1.10 [34 H, m, Pr^i and $(CH_3)_2CHCO$]; δ_C($CDCl_3$) 180.36 (Pr^i *C*=O), 158.75 (C-6), 153.56 (C-2), 152.31 (C-4), 145.39 (Ph, C-1), 142.53 (Ph, C-2), 138.44 (C-8), 134.71 (Ph, C-5), 125.73 (Ph, C-3), 125.16 (Ph, C-4 and -6), 119.44 (C-5), 87.77 (C-1′), 82.11 (C-4′), 80.32 (C-2′), 69.58 (C-3′), 59.60 (C-5′), 58.22 (OCH_3), 17.30 [$CH(CH_3)_2$], 35.55 [$COCH(CH_3)_2$], 18.99 [$COCH(CH_3)_2$] and 13.41, 12.73, 12.50 and 12.38 [$CH(CH_3)_2$]; FAB-MS *m/z* 732.2 [M + H]$^+$.

2-*N*-Isobutyryl-6-*O*-(2-nitrophenyl)-2′-*O*-allyl-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 6b

Compound **6b** was prepared from precursor **5b** (0.50 g, 0.72 mmol) in the same manner as described for compound **6a**. The crude product was purified by column chromatography on silica gel (12 g) and on elution with hexane–ethyl acetate (4:1 and 3:1 v/v). The *pure product* **6b** was obtained as a foam (0.47 g, 85.5%) of R_f 0.24 on TLC in hexane–ethyl acetate (3:1 v/v) (Found: C, 55.60; H, 6.96; N, 11.13. $C_{35}H_{52}N_6O_9Si_2$ requires C, 55.52; H, 6.94; N, 11.10%); ν_{max}(KBr)/cm^{-1} 2949 and 2871 (CH st), 1628 and 1578 (C=C, N=C) and 1532 (NO_2); δ_H($CDCl_3$) 9.68 (1 H, br s, NH), 8.11 (1 H, s, H-8), 7.96–7.27 (4 H, ArH), 6.12 (1 H, m, allyl CH), 6.05 (1 H, d, *J* 5.5, H-1′), 5.19 (2 H, m, allyl $=CH_2$), 4.61–4.20 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 3.78 (2 H, m, allyl CH_2), 2.16 (1 H, m, Pr^iCHCO) and 1.07 [34 H, m, $(CH_3)_2$-CHCO and Pr^i]; δ_C($CDCl_3$) 180.49 (Pr^iCH C=O), 159.39 (C-6), 152.85 (C-2), 152.14 (C-4), 145.10 (Ph, C-1), 142.14 (Ph, C-2), 141.75 (C-8), 134.86 (Ph, C-5), 133.70 (allyl CH), 126.65 (Ph, C-3), 125.55 (Ph, C-4 and -6), 120.54 (C-5), 117.42 (allyl $=CH_2$), 88.45 (C-1′), 81.58 (C-4′), 81.15 (C-2′), 71.55 (allyl OCH_2), 68.58 (C-3′), 59.51 (C-5′), 35.50 [$(CH_3)_2CHCO$], 18.85 [$COCH(CH_3)_2$], 17.10 and 16.81 [$CH(CH_3)_2$] and 13.24, 12.74 and 12.37 [$(CH_3)_2CH$]; FAB-MS *m/z* 758.2 [M + H]$^+$.

2-*N*-Isobutyryl-6-*O*-(2-nitrophenyl)-2′-*O*-propargyl-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 6c

Compound **6c** was prepared from precursor **5c** (1.00 g, 1.46 mmol) in the same manner as described for compound **6a**. The crude product was purified by column chromatography on silica gel (20 g) and elution with hexane–ethyl acetate (4:1 v/v). The *pure product* **6c** was obtained as a foam (0.88 g, 80.0%) of R_f 0.26 on TLC in hexane–ethyl acetate (3:1 v/v) (Found: C, 55.74; H, 6.71; N, 11.11. $C_{35}H_{50}N_6O_9Si_2$ requires C, 55.69; H, 6.69; N, 11.13%); ν_{max}(KBr)/cm^{-1} 3336 (propargyl C≡C–H), 2948 and 2868 (CH st), 1630 and 1578 (C=C, N=C) and 1532 (NO_2); δ_H-($CDCl_3$) 10.61 (1 H, br s, NH), 8.13 (1 H, s, H-8), 7.96–7.24 (4 H, ArH), 6.03 (1 H, d, *J* 5.4, H-1′), 4.73 (2 H, AB, *J* 16, propargyl CH_2), 4.62–4.19 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 2.57 (1 H, s, propargyl CH), 2.18 [1 H, m, $(CH_3)_2CHCO$] and 1.11 [34 H, m, Pr^i and $(CH_3)_2CHCO$]; δ_C($CDCl_3$) 181.00 (Pr^iC=O), 158.70 (C-6), 154.17 (C-2), 152.00 (C-4), 145.40 (Ph, C-1), 142.51 (Ph, C-2), 138.53 (C-8), 134.40 (Ph, C-5), 126.22 (Ph, C-3), 125.45 (Ph, C-4 and -6), 118.61 (C-5), 88.21 (C-1′), 81.19 (C-4′), 79.87 (C-2′), 79.10 (propargyl q C), 75.22 (propargyl CH), 69.61 (C-3′), 59.65 (C-5′), 58.13 (propargyl CH_2), 35.49 [$(CH_3)_2$-CHCO], 18.90 [$(CH_3)_2CHCO$], 17.26 [$(CH_3)_2CH$] and 13.20, 12.87 and 12.41 [$(CH_3)_2CH$]; FAB-MS *m/z* 756.2 [M + H]$^+$.

2′-*O*-Cyanomethyl-2-*N*-isobutyryl-6-*O*-(2-nitrophenyl)-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 6d

Compound **6d** was prepared from precursor **5d** (1.00 g, 1.46 mmol) in the same manner as described for compound **6a**. The crude product was purified by column chromatography on silica gel (25 g) and elution with hexane–ethyl acetate (4:1 and 3:1 v/v). The pure product **6d** was obtained as a foam (0.84 g, 76.1%) of R_f 0.20 on TLC in hexane–ethyl acetate (3:1 v/v) (Found: C, 54.09; H, 6.58; N, 13.03. $C_{34}H_{49}N_7O_9Si_2$ requires C, 54.01; H, 6.55; N, 12.97%); ν_{max}(KBr)/cm^{-1} 2947 and 2869 (CH st), 2180 (cyanomethyl CN), 1628 and 1577 (C=C, N=C) and 1532 (NO_2); δ_H($CDCl_3$) 9.71 (1 H, br s, NH), 8.13 (1 H, s, H-8), 7.96–7.24 (4 H, ArH), 6.03 (1 H, d, *J* 5.4, H-1′), 4.61–4.19 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 4.25 (2 H, AB, *J* 15.5, CH_2CN), 2.24 [1 H, m, $(CH_3)_2CHCO$] and 1.09 [34 H, m, Pr^i

and (CH_3)$_2$CHCO]; δ_C($CDCl_3$) 180.91 (Pr^i*C*=O), 158.90 (C-6), 153.81 (C-2), 152.58 (C-4), 145.21 (Ph, C-1), 142.35 (Ph, C-2), 138.21 (C-8), 134.49 (Ph, C-5), 126.11 (Ph, C-3), 125.37 (Ph, C-4 and -6), 115.71 (CH_2*C*N), 118.39 (C-5), 87.71 (C-1′), 81.65 (C-4′), 81.00 (C-2′), 69.58 (C-3′), 59.48 (C-5′), 56.25 (*C*H_2CN), 35.27 [(CH_3)$_2$*C*HCO], 18.88 [(*C*H_3)$_2$CHCO], 17.11 [(*C*H_3)$_2$CH], 13.35, 12.79 and 12.40 [(CH_3)$_2$*C*H]; ES-MS *m/z* 756.2 (M^+).

2′-*O*-(Ethoxycarbonyl)methyl-2-*N*-isobutyryl-6-*O*-(2-nitrophenyl)-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 6e

Compound **6e** was prepared from precursor **5e** (1.00 g, 1.36 mmol) in the same manner as described for compound **6a**. The crude product was purified by column chromatography on silica gel (25 g) and elution with hexane–ethyl acetate (4:1 and 3:1 v/v). The *pure product* **6e** was obtained as a foam (0.88 g, 80.6%) of R_f 0.18 on TLC in hexane–ethyl acetate (3:1 v/v) (Found: C, 53.77; H, 6.79; N, 10.48. $C_{36}H_{54}N_6O_{11}Si_2$ requires C, 53.70; H, 6.77; N, 10.44%); ν_{max}(KBr)/cm^{-1} 2947 and 2869 (CH st), 1730 ([ethoxycarbonyl]methyl C=O), 1629 and 1576 (C=C, N=C) and 1531 (NO_2); δ_H($CDCl_3$) 9.71 (1 H, br s, NH), 8.13 (1 H, s, H-8), 7.96–7.24 (4 H, ArH), 6.03 (1 H, d, *J* 5.4, H-1′), 4.61–4.19 (5 H, m, H-2′, -3′, -4′, H_2-5′), 4.24 (2 H, AB, *J* 16, [ethoxycarbonyl]-methyl CH_2), 3.40 (2 H, m, [ethoxycarbonyl]methyl CH_2), 1.79 (3 H, m, [ethoxycarbonyl]methyl CH_3), 2.13 [1 H, m, (CH_3)$_2$-C*H*CO] and 1.11 [34 H, m, (CH_3)$_2$CHCO and Pr^i]; δ_C($CDCl_3$) 180.41 (Pr^i, *C*=O), 169.59 ([ethoxycarbonyl]methyl C=O), 158.69 (C-6), 153.98 (C-2), 152.51 (C-4), 145.30 (Ph, C-1), 142.41 (Ph, C-2), 138.59 (C-8), 134.31 (Ph, C-5), 125.82 (Ph, C-3), 125.21 (Ph, C-4 and -6), 119.51 (C-5), 87.83 (C-1′), 82.00 (C-4′), 80.61 (C-2′), 69.62 (C-3′), 68.19 ([ethoxycarbonyl]methyl CH_2), 60.65 ([ethoxycarbonyl]methyl CH_2), 59.65 (C-5′), 35.61 [(CH_3)$_2$-*C*HCO], 18.91 [(*C*H_3)$_2$CHCO], 17.21 [(*C*H_3)$_2$CH], 13.20, 12.81, 12.54 and 12.49 [(CH_3)$_2$*C*H] and 12.83 ([ethoxycarbonyl]methyl CH_3); ES-MS *m/z* 803.2 (M^+).

2-*N*-Isobutyryl-2′-*O*-methyl-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 7a

Compound **6a** (0.75 g, 1.03 mmol) was dried by evaporation of dry acetonitrile (10 cm^3) and was dissolved in anhydrous acetonitrile (10 cm^3) under argon. A mixture of *N*,*N*,*N*′,*N*′-tetramethylguanidine (0.4 cm^3, 3.19 mmol) and 2-nitrobenzaldehyde oxime (0.55 g, 3.31 mmol) in acetonitrile (5 cm^3) was added to the stirred mixture with exclusion of moisture. The reaction mixture was left at room temperature for 16 h. TLC showed more or less complete reaction. The solvent was evaporated off *in vacuo* and the residue was dissolved in ethyl acetate (50 cm^3), the solution was washed twice with aq. sodium hydrogen carbonate (50 cm^3; 1 mol dm^{-3}), and the organic layer was separated, dried (Na_2SO_4), filtered, and evaporated *in vacuo*. The *crude product* **7a** was purified by column chromatography on silica gel (15 g) and elution with hexane–ethyl acetate (2:1 and 1:1 v/v). The pure product was obtained as a foam (0.56 g, 82.4%) of R_f 0.14 on TLC in hexane–ethyl acetate (1:1 v/v) (Found: C, 53.21; H, 7.81; N, 11.52. $C_{27}H_{47}N_5O_7Si_2$ requires C, 53.16; H, 7.78; N, 11.48%); ν_{max}(KBr)/cm^{-1} 2947 and 2869 (CH st), 1710 (C=O) and 1630 and 1578 (C=C, N=C); δ_H($CDCl_3$) 10.08 (1 H, br s, NH), 9.71 (1 H, br s, NH), 8.13 (1 H, s, H-8), 6.03 (1 H, d, *J* 5.4, H-1′), 4.57–4.23 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 3.41 (3 H, s, OCH_3), 2.12 [1 H, m, (CH_3)$_2$C*H*CO] and 1.07 [34 H, m, (CH_3)$_2$CHCO and Pr^i]; δ_C($CDCl_3$) 180.34 (Pr^i*C*=O), 156.75 (C-6), 148.88 (C-2), 147.59 (C-4), 137.32 (C-8), 112.43 (C-5), 87.87 (C-1′), 82.43 (C-4′), 80.34 (C-2′), 69.65 (C-3′), 59.61 (C-5′), 58.03 (OCH_3), 35.67 [(CH_3)$_2$*C*HCO], 18.80 [(*C*H_3)$_2$CHCO], 17.32–16.66 [(*C*H_3)$_2$CH] and 13.31, 12.73, 12.60 and 12.47 [(CH_3)$_2$*C*H]; FAB-MS *m/z* 611.1 [M + H]$^+$.

2′-*O*-Allyl-2-*N*-isobutyryl-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 7b

Compound **7b** was prepared from precursor **6b** (0.40 g, 0.53 mmol) in the same manner as described for compound **7a**. The crude product was purified by column chromatography on silica gel (8 g) and elution with hexane–ethyl acetate (1:1 v/v). The *pure product* **7b** was obtained as a foam (0.26 g, 76.2%) of R_f 0.23 on TLC in hexane–ethyl acetate (1:1 v/v) (Found: C, 54.85; H, 7.80; N, 11.05. $C_{29}H_{49}N_5O_7Si_2$ requires C, 54.76; H, 7.78; N, 11.01%); ν_{max}(KBr)/cm^{-1} 2947 and 2869 (CH st), 1711 (C=O) and 1627 and 1574 (C=C, N=C); δ_H($CDCl_3$) 10.12 (1 H, br s, NH), 9.68 (1 H, br s, NH), 8.11 (1 H, s, H-8), 6.09 (1 H, m, allyl CH), 6.05 (1 H, d, *J* 5.5, H-1′), 5.17 (2 H, m, allyl =CH_2), 4.61–4.17 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 3.81 (2 H, m, allyl CH_2), 2.19 [1 H, m, (CH_3)$_2$C*H*CO] and 1.07 [34 H, m, (CH_3)$_2$CHCO and Pr^i]; δ_C($CDCl_3$) 180.34 (Pr^i*C*=O), 157.11 (C-6), 148.56 (C-2), 147.19 (C-4), 137.75 (C-8), 133.93 (allyl CH), 121.15 (C-5), 116.77 (allyl =CH_2), 87.68 (C-1′), 81.27 (C-2′ and -4′), 71.05 (allyl OCH_2), 68.57 (C-3′), 59.51 (C-5′), 35.64 [(CH_3)$_2$CHCO], 18.81 [(*C*H_3)$_2$CHCO] and 17.07–16.77 [(*C*H_3)$_2$CH] and 13.94, 12.77, 12.58 and 12.35 [(CH_3)$_2$*C*H]; FAB-MS *m/z* 637.1 [M + H]$^+$.

2-*N*-Isobutyryl-2′-*O*-propargyl-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 7c

Compound **7c** was prepared from precursor **6c** (0.75 g, 0.99 mmol) in the same manner as described for compound **7a**. The crude product was purified by column chromatography on silica gel (15 g) and eluted with hexane–ethyl acetate (2:1 v/v). The *pure product* **7c** was obtained as a foam (0.51 g, 80.9%) of R_f 0.25 on TLC in hexane–ethyl acetate (1:1 v/v) (Found: C, 55.09; H, 7.53; N, 11.10. $C_{29}H_{47}N_5O_7Si_2$ requires C, 54.94; H, 7.49; N, 11.05%); ν_{max}(KBr)/cm^{-1} 3340 (propargyl C≡CH), 2949 and 2867 (CH st), 2150 (propargyl C≡CH), 1711 (C=O) and 1630 and 1576 (C=C, N=C); δ_H($CDCl_3$) 10.23 (1 H, br s, NH), 9.71 (1 H, br s, NH), 8.10 (1 H, s, H-8), 6.00 (1 H, d, *J* 5.4, H-1′), 4.73 (2 H, AB system, *J* 15.8, propargyl CH_2), 4.57–4.21 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 2.55 (1 H, s, propargyl CH), 2.11 [1 H, m, (CH_3)$_2$C*H*CO] and 1.10 [34 H, m, (CH_3)$_2$CH]; δ_C($CDCl_3$) 181.00 (Pr^i*C*=O), 157.05 (C-6), 155.70 (C-2), 149.17 (C-4), 136.51 (C-8), 120.61 (C-5), 87.21 (C-1′), 81.24 (C-4′), 80.87 (C-2′), 79.15 (propargyl q, C), 75.29 (propargyl CH), 68.51 (C-3′), 59.55 (C-5′), 58.23 (propargyl CH_2), 35.46 [(CH_3)$_2$CHCO], 18.98 [(*C*H_3)$_2$CHCO], 17.26 [(*C*H_3)$_2$CH] and 13.20, 12.87 and 12.41 [(CH_3)$_2$*C*H]; FAB-MS *m/z* 635.1 [M + H]$^+$.

2-*N*-Isobutyryl-2′-*O*-cyanomethyl-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 7d

Compound **7d** was prepared from precursor **6d** (1.00 g, 1.46 mmol) in the same manner as described for compound **7a**. The crude product was purified by column chromatography on silica gel (25 g) and elution with hexane–ethyl acetate (4:1 and 3:1 v/v). The *pure product* **7d** was obtained as a foam (0.84 g, 76.1%) of R_f 0.21 on TLC in hexane–ethyl acetate (1:1 v/v) (Found: C, 53.07; H, 7.35; N, 13.29. $C_{28}H_{46}N_6O_7Si_2$ requires C, 52.96; H, 7.32; N, 13.24%); ν_{max}(KBr)/cm^{-1} 2947 and 2866 (CH st), 2175 (cyanomethyl CN), 1712 (C=O) and 1630 and 1576 (C=C, N=C); δ_H($CDCl_3$) 10.29 (1 H, br s, NH), 9.71 (1 H, br s, NH), 8.10 (1 H, s, H-8), 6.01 (1 H, d, *J* 5.5, H-1′), 4.56–4.23 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 4.28 (2 H, AB, *J* 15.5, CH_2CN), 2.21 [1 H, m, (CH_3)$_2$C*H*CO] and 1.13 [34 H, m, (CH_3)$_2$CHCO and Pr^i]; δ_C($CDCl_3$) 180.91 (Pr^i*C*=O), 158.90 (C-6), 153.81 (C-2), 152.28 (C-4), 145.21 (Ph, C-1), 142.35 (C-2), 138.21 (C-8), 134.49 (Ph, C-2), 126.11 (Ph, C-4), 125.37 (Ph, C-3 and -5), 115.71 (CH_2*C*N), 118.39 (C-5), 87.71 (C-1′), 81.65 (C-4′), 81.00 (C-2′), 69.58 (C-3′), 59.48 (C-5′), 56.25 (*C*H_2CN), 35.27 [(CH_3)$_2$*C*HCO], 18.88 [(*C*H_3)$_2$CHCO], 17.11 [(*C*H_3)$_2$CH] and 13.35, 12.79 and 12.40 [(CH_3)$_2$*C*Hs]; ES-MS *m/z* 635.1 (M^+).

2-*N*-Isobutyryl-2′-*O*-(ethoxycarbonyl)methyl-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 7e

Compound **7e** was prepared from precursor **6e** (0.75 g, 0.93 mmol) in the same manner as described for compound **7a**. The

crude product was purified by column chromatography on silica gel (15 g) and elution with hexane–ethyl acetate (2:1 and 1:1 v/v). The *pure product* **7e** was obtained as a foam (0.56 g, 82.4%) of R_f 0.20 on TLC in hexane–ethyl acetate (1:1 v/v) (Found: C, 52.89; H, 7.63; N, 10.31. $C_{30}H_{51}N_5O_9Si_2$ requires C, 52.83; H, 7.55; N, 10.27%); ν_{max}(KBr)/cm^{-1} 2948 and 2867 (CH st), 1725 and 1712 (C=O) and 1629 and 1575 (C=C, N=C); δ_H($CDCl_3$) 10.19 (1 H, br s, NH), 9.71 (1 H, br s, NH), 8.16 (1 H, s, H-8), 6.09 (1 H, d, *J* 5.4, H-1′), 4.65–4.23 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 4.22 (2 H, AB, *J* 16, [ethoxycarbonyl]methyl CH_2), 3.34 (2 H, m, [ethoxycarbonyl]methyl CH_2), 1.75 (3 H, m, CH_2CO_2-CH_2CH_3), 2.18 [1 H, m, $(CH_3)_2CHCO$] and 1.10 [34 H, m, $(CH_3)_2CHCO$ and Pr^i]; δ_C($CDCl_3$) 180.21 (Pr^i*C*=O), 169.51 (CH_2CO_2Et), 156.89 (C-6), 148.98 (C-2), 147.51 (C-4), 137.11 (C-8), 112.61 (C-5), 87.70 (C-1′), 82.22 (C-4′), 80.11 (C-2′), 69.70 (C-3′), 68.29 ([ethoxycarbonyl]methyl CH_2) 60.61 ([ethoxycarbonyl]methyl CH_2), 59.61 (C-5′), 35.55 [$(CH_3)_2CHCO$], 18.70 [$(CH_3)_2CHCO$], 17.22–16.79 [$(CH_3)_2CH$], 13.26, 12.89, 12.63 and 12.54 [$(CH_3)_2CH$] and 12.71 ($CH_2CO_2CH_2CH_3$); ES-MS *m/z* 682.2 (M^+).

6-*O*-(*tert*-Butyldiphenylsilyl)-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)guanosine 8

Alternative 1. Guanosine (10.05 g, 35.45 mmol) was dried by evaporation of anhydrous pyridine and was then mixed with pyridine–DMF (200 cm^3; 1:4 v/v) under argon. TIPDSCl (12.2 cm^3, 38.91 mmol) was added to the stirred mixture with exclusion of moisture. After 1 h at 50 °C TLC showed complete reaction. The reaction was quenched with methanol (5 cm^3) and the solvent was evaporated off *in vacuo*. The residual syrup was coevaporated with toluene (2 × 80 cm^3) to leave a white–yellow foam.

Alternative 2. Guanosine (5.00 g, 17.65 mmol) was dried by evaporation of dry pyridine (40 cm^3) and was then dissolved in dry DMF (80 cm^3) under argon. Imidazole (7.3 g, 107.20 mmol) and TIPDSCl (9.0 cm^3, 28.70 mmol) were added to the stirred mixture with exclusion of moisture. The mixture was left at room temperature. TLC showed complete reaction after 2 h. The reaction mixture was concentrated under reduced pressure and purified by silica chromatography (175 g) and elution with a gradient of ethanol (5–10%) in dichloromethane. The pure product was obtained as a foam (7.89 g, 85.0%). The crude product from above was dissolved in dry dichloromethane (300 cm^3) under argon. DMAP (0.9 g, 7.37 mmol), triethylamine (25 cm^3, 179.51 mmol) and TBDPSCl (11 cm^3, 42.30 mmol) were added to the stirred mixture with exclusion of moisture. The mixture was left at room temperature for 8 h. TLC (ethanol–dichloromethane 5:95 v/v) showed complete reaction. Dichloromethane was added (200 cm^3) and the solution was washed successively with 0.5 M aq. sodium phosphate buffer, pH 7 (500 cm^3) and saturated brine (500 cm^3). The organic layer was dried (Na_2SO_4), then filtered, and the solvent was removed *in vacuo.*

2-*N*-Dimethylaminomethylene-2′-*O*-methylguanosine 10a

Compound **8** (6.6 g, 8.63 mmol) was dried by evaporation of anhydrous acetonitrile (50 cm^3) and was then dissolved in anhydrous acetonitrile (75 cm^3) under argon. The solution was cooled in an ice-bath. Methyl iodide (3 cm^3, 48.19 mmol) and BEMP (5.1 cm^3, 17.62 mmol) were added to the stirred mixture with exclusion of moisture. The solution was kept for 10 min at 0 °C and was then stirred for 1.5 h at room temperature. TLC showed more or less complete reaction. Solvent was removed *in vacuo* to leave compound **9a** as a foam. Ethyl acetate (200 cm^3) was added and the solution was washed with 0.5 mol dm^{-3} aq. sodium phosphate buffer, pH 7 (200 cm^3). The organic layer was dried (Na_2SO_4), filtered, and evaporated *in vacuo*. The residue was dissolved in dry THF (75 cm^3) and 1 mol dm^{-3} TBAF in THF (10 cm^3) was added to the stirred solution. TLC showed complete reaction after 15 min. The reaction mixture was quenched with pyridine–methanol–water (10 cm^3; 3:1:1 by volume), and the solution was poured into a stirred suspension of the pyridinium form of Dowex 50 W x4-200 resin (40 g) in pyridine–methanol–water (20 cm^3, 3:1:1 by volume). The mixture was stirred for 30 min and the resin was filtered off, and washed with the above pyridine–methanol–water solution (3 × 25 cm^3). Combined filtrate and washings were evaporated to dryness *in vacuo*, and residual pyridine was removed by addition and evaporation of toluene. The resulting foam was dissolved in dry methanol (100 cm^3) under argon and dimethylformamide dimethyl acetal (12 cm^3, 90.02 mmol) was added. The mixture was stirred overnight at room temperature. TLC showed complete reaction. The reaction mixture was evaporated *in vacuo* and the residue was dried by evaporation of toluene (2 × 30 cm^3). The crude product was purified by column chromatography on silica gel (80 g) and elution with a gradient of ethanol 5–20% in dichloromethane. Pure title compound **10a** was obtained as a solid foam (1.49 g, 49.0% calculated from guanosine) of R_f 0.15 on TLC in ethanol–dichloromethane (1:4 v/v) (Found: C, 47.74; H, 5.73; N, 23.87. Calc. for $C_{14}H_{20}N_6O_5$: C, 47.71; H, 5.73; N, 23.85%); ν_{max}(KBr)/cm^{-1} 3436br (OH), 1713 (C=O) and 1628 and 1576 (C=C, N=C); δ_H[$(CD_3)_2SO$] 9.98 (1 H, br s, NH), 8.59 (1 H, s, C*H*=N), 8.17 (1 H, s, H-8), 6.00 (1 H, d, *J* 5.4, H-1′), 4.55–4.25 (5 H, m, H-2′, -3′, -4′, H_2-5′), 3.31 (3 H, s, OCH_3), 3.21 (3 H, s, amidine CH_3) and 3.11 (3 H, s, amidine CH_3). The ^{13}C NMR data were in agreement with previously published data.[17] ES-MS *m/z* 352.6 (M^+).

2′-*O*-Allyl-2-*N*-dimethylaminomethyleneguanosine 10b

Compound **8** (7 g, 9.16 mmol) was treated with allyl bromide (1.6 cm^3, 18.49 mmol) and BEMP (5.5 cm^3, 19.01 mmol) according to the procedure used to prepare compound **10a** above; the reaction time was extended to 2 h. Desilylation and protection of the 2-amino group of the intermediate **9b** were performed as described for the preparation of compound **10a**. The crude product was purified by column chromatography on silica gel (80 g) and elution with a gradient of ethanol 5–20% in dichloromethane. Pure title compound **10b** was obtained as a solid foam (1.77 g, 51.1% calculated from guanosine) of R_f 0.27 on TLC in ethanol–dichloromethane (1:4 v/v) (Found: C, 50.62; H, 6.13; N, 22.15. Calc. for $C_{16}H_{22}N_6O_5$: C, 50.51; H, 6.11; N, 22.10%); ν_{max}(KBr)/cm^{-1} 3438br (OH), 2947 and 2868 (CH st), 1711 (carbamoyl C=O) and 1626 and 1577 (C=C, N=C); δ_H[$(CD_3)_2SO$] 9.98 (1 H, br s, NH), 8.49 (1 H, s, C*H*=N), 8.15 (1 H, s, H-8), 6.13 (1 H, m, allyl CH), 6.07 (1 H, d, *J* 5.5, H-1′), 5.21 (2 H, m, allyl =CH_2), 4.55–4.27 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 3.78 (2 H, m, allyl CH_2), 3.12 (3 H, s, amidine CH_3) and 3.01 (3 H, s, amidine CH_3). The ^{13}C NMR data were in agreement with previously published data.[11] ES-MS *m/z* 379.6 (M^+).

2-*N*-Dimethylaminomethylene-2′-*O*-propargylguanosine 10c

Compound **8** (10.00 g, 13.09 mmol) was treated with propargyl bromide (7.3 cm^3, 65.28 mmol) and BEMP (9.4 cm^3, 32.48 mmol) according to the procedure for compound **10a**. Desilylation and protection of the 2-amino group in the intermediate **9c** were performed as described for the preparation of compound **10a**. The crude product was purified by column chromatography on silica gel (100 g) and elution with a gradient of ethanol 5–20% in dichloromethane. Pure *title compound* **10c** was obtained as a solid foam (2.53 g, 51.3% calculated from guanosine) of R_f 0.31 on TLC in ethanol–dichloromethane (1:4 v/v) (Found: C, 51.09; H, 5.39; N, 22.48. $C_{16}H_{20}N_6O_5$ requires C, 51.05; H, 5.37; N, 22.33%); ν_{max}(KBr)/cm^{-1} 3340br (OH), 3340 (propargyl C≡C–H), 1711 (C=O) and 1624 and 1575 (C=C, N=C); δ_H[$(CD_3)_2SO$] 10.01 (1 H, br s, NH), 8.51 (1 H, s, C*H*=N), 8.19 (1 H, s, H-8), 6.03 (1 H, d, *J* 5.5, H-1′), 4.73 (2 H, AB, *J* 16, propargyl CH_2), 4.64–4.23 (5 H, m, H-2′, -3′, -4′ and H_2-5′), 3.17 (3 H, s, amidine CH_3), 3.00 (3 H, s, amidine CH_3) and 2.59 (1 H, s, propargyl CH); δ_C[$(CD_3)_2SO$] 158.00 (amidine

CH) 157.54 and 157.37 (C-2 and -6), 149.82 (C-4), 136.68 (C-8), 119.77 (C-5), 85.90 (C-1′), 84.95 (C-4′), 80.10 (C-2′), 79.96 (propargyl q C), 77.46 (propargyl CH), 68.70 (C-3′), 61.15 (C-5′), 56.92 (propargyl CH_2) and 40.63 and 34.61 (amidine CH_3s); ES-MS *m/z* 376.7 (M^+).

2′-*O*-Cyanomethyl-2-*N*-dimethylaminomethyleneguanosine 10d

Compound **8** (6.00 g, 7.71 mmol) was treated with bromoacetonitrile (1.34 cm^3, 19.28 mmol) and BEMP (4.5 cm^3, 15.55 mmol) according to the procedure for compound **10a**. Desilylation and protection of the 2-amino group in intermediate **9d** were performed as described for the preparation of compound **10a**. The crude product was purified by column chromatography on silica gel (100 g) and elution with a gradient of ethanol 5–20% in dichloromethane. The title compound **10d** was obtained as a solid foam (1.38 g, 47.4% calculated from guanosine) of R_f 0.33 on TLC in ethanol–dichloromethane (1:4 v/v) (Found: C, 47.64; H, 5.11; N, 26.09. $C_{15}H_{19}N_7O_5$ requires C, 47.73; H, 5.08; N, 25.99%); ν_{max}(KBr)/cm^{-1} 3431br (OH), 2173 (cyanomethyl CN), 1710 (C=O) and 1625 and 1574 (C=C, N=C); δ_H[$(CD_3)_2SO$] 10.19 (1 H, br s, NH), 8.46 (1 H, s, C*H*=N), 8.16 (1 H, s, H-8), 6.08 (1 H, d, *J* 5.4, H-1′), 4.64–4.23 (5 H, m, H-2′, -3′, -4′, H_2-5′), 4.28 (2 H, AB, *J* 15.5, CH_2CN), 3.19 (3 H, s, amidine CH_3) and 3.08 (3 H, s, amidine CH_3); δ_C[$(CD_3)_2SO$] 158.40 (amidine CH), 157.96 and 156.83 (C-2 and -6), 150.03 (C-4), 136.14 (C-8), 119.88 (C-5), 116.33 (CH_2CN), 85.99 (C-1′), 83.01 (C-4′), 81.87 (C-2′), 69.13 (C-3′), 58.50 (C-5′), 56.06 (CH_2CN) and 40.99 and 34.82 (amidine CH_3s); ES-MS *m/z* 377.6 (M^+).

2-*N*-Dimethylaminomethylene-2′-*O*-[(ethoxycarbonyl)methyl]-guanosine 10e

Compound **8** (10.00 g, 13.09 mmol) was treated with ethyl bromoacetate (3 cm^3, 26.94 mmol) and BEMP (9.4 cm^3, 32.48 mmol) according to the procedure for compound **10a**. Desilylation and protection of the 2-amino group of intermediate **9e** were performed as described for the preparation of compound **10a** except that 1 mol dm^{-3} TBAF mixed with 1 equiv. acetic acid in THF (1:1 v/v) was used for the desilylation. The crude product was purified by column chromatography on silica gel (100 g) and elution with a gradient of ethanol 5–20% in dichloromethane. Pure *title compound* **10e** was obtained as a solid foam (2.70 g, 48.6% calculated from guanosine) of R_f 0.24 on TLC on ethanol–dichloromethane (1:4 v/v) (Found: C, 48.16; H, 5.73; N, 19.85. $C_{17}H_{24}N_6O_7$ requires C, 48.10; H, 5.71; N, 19.80%); ν_{max}(KBr)/cm^{-1} 3431br (OH), 1725 and 1713 (C=O) and 1626 and 1575 (C=C, N=C); δ_H[$(CD_3)_2SO$] 10.19 (1 H, br s, NH), 8.51 (1 H, s, C*H*=N), 8.16 (1 H, s, H-8), 6.09 (1 H, d, *J* 5.5, H-1′), 4.67–4.29 (5 H, m, H-2′, -3′, -4′, and H_2-5′), 4.21 (2 H, AB, *J* 16, [ethoxycarbonyl]methyl CH_2), 3.22 (3 H, s, amidine CH_3) and 3.09 (3 H, s, amidine CH_3); δ_C[$(CD_3)_2SO$] 169.79 ([ethoxycarbonyl]methyl C=O), 158.04 (amidine CH) 157.37 and 156.94 (C-2 and -6), 150.82 (C-4), 137.60 (C-8), 120.01 (C-5), 85.45 (C-1′), 84.36 (C-4′), 83.38 (C-2′), 69.83 (C-3′), 68.46 ([ethoxycarbonyl]methyl CH_2), 63.36 ([ethoxycarbonyl]-methyl CH_2), 61.35 (C-5′), 40.63 and 34.61 (amidine CH_3s) and 13.88 ($CH_2CO_2CH_2CH_3$); ES-MS *m/z* 424.6 (M^+).

2-Amino-2′-*O*-methylpurine-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)riboside 12

Compound **5a** (0.50 g, 0.75 mmol) was treated with 4 mol equiv. of hydrazine monohydrate at room temp. for 24 h following the procedure of Connolly.[27] The crude product was purified by column chromatography on silica gel (10 g) and elution with a gradient of ethanol (0–15%) in dichloromethane. The pure product, compound **11**, was obtained as a foam (300 mg, 72.0%).

The above material was converted into title compound **12** following the procedure of Connolly.[27] The crude product was purified by column chromatography on silica gel (10 g) and elution with a gradient of methanol (0–5%) in dichloromethane. The pure product was obtained as a foam (196 mg, 68.0%) of R_f 0.68 on TLC in ethanol–dichloromethane (1:9 v/v); λ_{max}(EtOH)/nm 310 (6800) and 246 (5800). Good elemental analytical data were not obtained probably due to traces of silver oxide; ν_{max}(KBr)/cm^{-1} 3322 (NH), 2947 and 2869 (CH st) and 1614 and 1580 (C=C, C=N); δ_H($CDCl_3$) 8.66 (1 H, s, H-8), 8.08 (1 H, s, H-6), 5.93 (1 H, s, H-1′), 5.35 (2 H, br s, NH_2), 4.57 (1 H, m, H-3′), 4.23–3.96 (4 H, m, H-2′, -4′ and H_2-5′), 3.67 (3 H, s, OCH_3) and 1.04 (28 H, m, Pr^i); δ_C($CDCl_3$) 159.79 (C-2), 152.08 (C-4), 149.90 (C-6), 140.36 (C-8), 128.81 (C-5), 87.51 (C-1′), 83.42 (C-4′), 81.10 (C-2′), 69.56 (C-3′), 59.76 (C-5′), 59.43 (OCH_3), 17.22, 16.99 and 16.81 [$(CH_3)_2CH$] and 13.41, 12.88 and 12.45 [$(CH_3)_2CH$]; ES-MS *m/z* 524.3 (M^+).

2,6-Diamino-2′-*O*-methylpurine-3′,5′-*O*-(tetraisopropyldisiloxane-1,3-diyl)riboside 13

Compound **5** (0.50 g, 0.75 mmol) was treated with a solution of ammonia (10 g) in THF (50 ml) following the procedure of Sproat *et al.*[11] The crude product was purified by column chromatography on silica gel (10 g) and elution with a gradient of methanol (0–10%) in dichloromethane. The pure product **13** was obtained as a foam (400 mg, 98.6%) of R_f 0.57 on TLC in ethanol–dichloromethane (1:9 v/v); λ_{max}(EtOH)/nm 258 (8900) and 282 (9700) (Found: C, 51.31; H, 7.89; N, 15.68. Calc. for $C_{23}H_{42}N_6O_5Si_2$: C, 51.26; H, 7.87; N, 15.60); ν_{max}(KBr)/cm^{-1} 3332 and 3193 (NH), 2946 and 2869 (CH st) and 1602 (C=C, C=N); δ_H($CDCl_3$) 7.82 (1 H, s, H-8), 6.01 (1 H, s, H-1′), 5.89 (2 H, br s, NH_2), 5.19 (2 H, br s, NH_2), 4.60 (1 H, m, H-3′), 4.23–3.97 (4 H, m, H-2′, -4′ and H_2-5′), 3.67 (3 H, s, OCH_3) and 1.09 (28 H, m, Pr^i). The ^{13}C NMR data were in agreement with previously published data.[11] ES-MS *m/z* 539.3 (M^+).

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