Nucleotides

Part LXIII[1])

New 2-(4-Nitrophenyl)ethyl(Npe)- and 2-(4-Nitrophenyl)ethoxycarbonyl(Npeoc)-Protected 2′-Deoxyribonucleosides and Their 3′-Phosphoramidites – Versatile Building Blocks for Oligonucleotide Synthesis

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With best personal wishes dedicated to Prof. Dr. *Frank Seela* on the occasion of his 60th birthday

A series of new base-protected and 5′-*O*-(4-monomethoxytrityl)- or 5′-*O*-(4,4′-dimethoxytrityl)-substituted 3′-(2-cyanoethyl diisopropylphosphoramidites) and 3′-[2-(4-nitrophenyl)ethyl diisopropylphosphoramidites] **52**–**66** and **67**–**82**, respectively, are prepared as potential building blocks for oligonucleotide synthesis (see *Scheme*). Thus, 3′,5′-di-*O*-acyl- and N^2,3′-*O*,5′-*O*-triacyl-2′-deoxyguanosines can easily be converted into the corresponding O^6-alkyl derivatives **6**, **8**, **10**, **12**, **14**, and **16** by a *Mitsunobu* reaction using the appropriate alcohol. Mild hydrolysis removes the acyl groups from the sugar moiety (→ **9**, **11**, **13**, **15**, and **19** (*via* **18**), resp.) which can then be tritylated (→ **38**–**42**) and phosphitylated (→ **57**–**61**) in the usual manner. N^2-[2-(4-nitrophenyl)ethoxycarbonyl]-substituted and N^2-[2-(4-nitrophenyl)ethoxycarbonyl]-O^6-[2-(4-nitrophenyl)ethyl]-substituted 2′-deoxyguanosines **5** and **7**, respectively, are synthesized as new starting materials for tritylation (→ **28**, **35**, and **37**) and phosphitylation (→ **54**, **56**, **70**, and **78**). Various O^4-alkylthymidines (see **20**–**24**) are also converted to their 5′-*O*-dimethoxytrityl derivatives (see **43**–**47)** and the corresponding phosphoramidites (see **62**–**66** and **79**–**82**).

1. Introduction. – Blocking groups [2] play a crucial role in synthetic approaches using polyfunctional molecules such as nucleosides and nucleotides. A broad variety of protecting strategies [3–7] have been recommended for the buildup of oligonucleotides which can nowadays easily be synthesized by machine-aided methods on solid-support materials [8–11]. Despite the fact that the use of acid- and base-labile protecting groups give very good results, we stressed for many years the idea of applying the 2-(4-nitrophenyl)ethyl (npe) and the 2-(4-nitrophenyl)ethoxycarbonyl (npeoc) group [12–14] as a versatile alternative providing an uniform protection of the amino, amide, hydroxy, mercapto, carboxy, and phosphate functions and the advantage of simultaneous cleavage under aprotic conditions by a β-elimination process [13]. Over the years, a large number of new npe- and npeoc-protected 2′-deoxyribonucleosides and their corresponding 3′-phosphoramidites have been synthesized and proven to be valuable intermediates for oligonucleotide synthesis. Furthermore, the synthesis of similar O^4-alkylthymidine and O^6-alkyl-2′-deoxyguanosine derivatives have been included in these studies. We will report in this paper about the syntheses and the

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characterizations of these building blocks starting from thymidine (**1**) and 2′-deoxyguanosine (**2**).

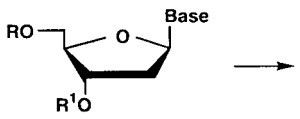
2. Syntheses. – Suitable protection of 2′-deoxyguanosine (**2**) can be seen as the most crucial presupposition for an effective and homogenous synthesis of oligo-2′-deoxyribonucleotides. Thus, 2′-deoxy-N^2-[2-(4-nitrophenyl)ethoxycarbonyl]guanosine (**5**) was synthesized by the transient protection method [14] using trimethylsilyl chloride for intermediary blocking of the sugar OH groups, followed by acylation with 2-(4-nitrophenyl)ethyl carbonochloridate (= 2-(4-nitrophenyl)ethyl chloroformate) (*cf.* the corresponding **3** and **4**) [12]. The preparation of 2′-deoxy-O^6-[2-(4-nitrophenyl)ethyl]-N^2-[2-(4-nitrophenyl)ethoxycarbonyl]guanosine (**7**) could be achieved in a one-pot reaction starting from 3′, 5′-di-*O*-acetyl-2′-deoxyguanosine [15] which was first subject to a *Mitsunobu* reaction [16] leading, under O^6-alkylation, to 3′,5′-di-*O*-acetyl-2′-deoxy-O^6-[2-(4-nitrophenyl)ethyl]guanosine. Subsequent acylation with 2-(4-nitrophenyl)ethyl carbonochloridate, followed by treatment with ammonia, led to **7** in an overall yield of 66% (*cf.* also **6**). In a similar manner, 2′-deoxy-O^6-methyl-N^2-[2-(4-nitrophenyl)ethoxycarbonyl]guanosine (**19**) was synthesized *via* its 3′,5′-di-*O*-acetyl derivative **18**, but in this case, besides O^6-methylation (→ **16**) also N^1-methylation to **17** took place in a substantial amount. Nevertheless, the *Mitsunobu* alkylation [17] is superior to nucleophilic displacement reactions of the activated amide function in **2** [18][19] and works also very well with 2′-deoxy-N^2,3′-*O*,5′-*O*-triisobutyrylguanosine [12][20] leading with MeOH, EtOH, ^{i}PrOH, and BuOH to the O^6-methyl-, O^6-ethyl-, O^6-isopropyl-, and O^6-butyl derivative **8**, **10**, **12**, and **14**, respectively, of which **10** and **12** were isolated as intermediates before hydrolysis to **11** and **13**, whereas **8** and **14** were converted without isolation into 2′-deoxy-N^2-isobutyryl-O^6-methylguanosine (**9**) and the corresponding O^6-butyl derivative **15**.

In the thymidine series, the *Mitsunobu* reaction leads, unfortunately, to alkylation at N(3) so that the synthesis of the O^4-alkyl derivatives **20**–**24** was achieved either by reaction with alkyl halide/silver carbonate [21][22] or *via* the triazolide method [22–25] and the use of O^4-[(2,4,6-triisopropylphenyl)sulfonyl] intermediates [26].

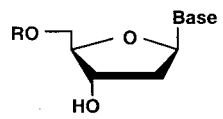
Tritylation reactions with 4-monomethoxy- and 4,4′-dimethoxytrityl chloride, respectively, proceeded very well by the conventional method and led, under selective substitution at the primary 5′-OH group, to **28** and **33**–**47** in yields > 85% (see also **25** [15], **26** and **27** [12], and **29**–**31** [15]). The synthesis by different routes of some of these compounds, *i.e.* **38**, **39**, **41**, and **43**–**46**, have already been described [26–29].

Phosphitylations were also performed by conventional methods applying either 2-cyanoethyl diisopropylphosphoramidochloridite (= chloro(2-cyanoethoxy)(diisopropylamino)phosphane; **48**) [30][31] with *Hünig*'s base as acid scavenger or with 2-cyanoethyl tetraisopropylphosphorodiamidite (= (2-cyanoethoxy)bis(diisopropylamino)phosphane; **49**) [32–35] and 1*H*-tetrazole in CH_2Cl_2 to convert the 5′-*O*-(4,4′-dimethoxytrityl)-2′-deoxyribonucleosides **33**–**47** into the corresponding 3′-(2-cyanoethyl diisopropylphosphoramidites) **52**–**66**. In a second series of reactions, we evaluated the 2-(4-nitrophenyl)ethyl group as a phosphate protecting group [36–38] and synthesized, from **25**–**36, 43**–**45**, and **47**, with the phosphitylating agents 2-(4-nitrophenyl)ethyl diisopropylphosphoramidochloridite (= chloro(diisopropylamino)-[2-(4-nitrophenyl)ethoxy]phosphane; **50**) [39] and 2-(4-nitrophenyl)ethyl tetraisopro-

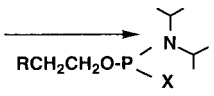
Scheme



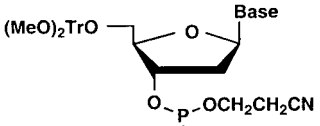
	Base	R	R1
1	Thy	H	H
2	Gua	H	H
3	Ade^{npeoc}	H	H
4	Cyt^{npeoc}	H	H
5	Gua^{npeoc}	H	H
6	Gua$^{npe^6}_{ibu}$	H	H
7	Gua$^{npe^6}_{npeoc}$	H	H
8	Gua$^{me^6}_{ibu}$	ibu	ibu
9	Gua$^{me^6}_{ibu}$	H	H
10	Gua$^{et^6}_{ibu}$	ibu	ibu
11	Gua$^{et^6}_{ibu}$	H	H
12	Gua$^{ipr^6}_{ibu}$	ibu	ibu
13	Gua$^{ipr^6}_{ibu}$	H	H
14	Gua$^{nbu^6}_{ibu}$	ibu	ibu
15	Gua$^{nbu^6}_{ibu}$	H	H
16	Gua$^{me^6}$	Ac	Ac
17	Gua$^{me^1}$	Ac	Ac
18	Gua$^{me^6}_{npeoc}$	Ac	Ac
19	Gua$^{me^6}_{npeoc}$	H	H
20	Thy$^{me^4}$	H	H
21	Thy$^{et^4}$	H	H
22	Thy$^{ipr^4}$	H	H
23	Thy$^{nbu^4}$	H	H
24	Thy$^{npe^4}$	H	H



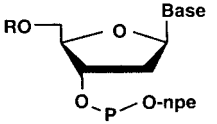
	Base	R
25	Thy	MeOTr
26	Ade^{npeoc}	MeOTr
27	Cyt^{npeoc}	MeOTr
28	Gua$^{npe^6}_{npeoc}$	MeOTr
29	Thy	$(MeO)_2Tr$
30	Ade^{bz}	$(MeO)_2Tr$
31	Cyt^{bz}	$(MeO)_2Tr$
32	Gua^{ibu}	$(MeO)_2Tr$
33	Ade^{npeoc}	$(MeO)_2Tr$
34	Cyt^{npeoc}	$(MeO)_2Tr$
35	Gua^{npeoc}	$(MeO)_2Tr$
36	Gua$^{npe^6}_{ibu}$	$(MeO)_2Tr$
37	Gua$^{npe^6}_{npeoc}$	$(MeO)_2Tr$
38	Gua$^{me^6}_{ibu}$	$(MeO)_2Tr$
39	Gua$^{et^6}_{ibu}$	$(MeO)_2Tr$
40	Gua$^{ipr^6}_{ibu}$	$(MeO)_2Tr$
41	Gua$^{nbu^6}_{ibu}$	$(MeO)_2Tr$
42	Gua$^{me^6}_{npeoc}$	$(MeO)_2Tr$
43	Thy$^{me^4}$	$(MeO)_2Tr$
44	Thy$^{et^4}$	$(MeO)_2Tr$
45	Thy$^{ipr^4}$	$(MeO)_2Tr$
46	Thy$^{nbu^4}$	$(MeO)_2Tr$
47	Thy$^{npe^4}$	$(MeO)_2Tr$



	R	X
48	CN	Cl
49	CN	$N(ipr)_2$
50	O_2N-C$_6$H$_4$-	Cl
51	O_2N-C$_6$H$_4$-	$N(ipr)_2$



	Base
52	Ade^{npeoc}
53	Cyt^{npeoc}
54	Gua^{npeoc}
55	Gua$^{npe^6}_{ibu}$
56	Gua$^{npe^6}_{npeoc}$
57	Gua$^{me^6}_{ibu}$
58	Gua$^{et^6}_{ibu}$
59	Gua$^{ipr^6}_{ibu}$
60	Gua$^{nbu^6}_{ibu}$
61	Gua$^{me^6}_{npeoc}$
62	Thy$^{me^4}$
63	Thy$^{et^4}$
64	Thy$^{ipr^4}$
65	Thy$^{nbu^4}$
66	Thy$^{npe^4}$



	Base	R
67	Thy	MeOTr
68	Ade^{npeoc}	MeOTr
69	Cyt^{npeoc}	MeOTr
70	Gua$^{npe^6}_{npeoc}$	MeOTr
71	Thy	$(MeO)_2Tr$
72	Ade^{bz}	$(MeO)_2Tr$
73	Cyt^{bz}	$(MeO)_2Tr$
74	Gua^{ibu}	$(MeO)_2Tr$
75	Ade^{npeoc}	$(MeO)_2Tr$
76	Cyt^{npeoc}	$(MeO)_2Tr$
77	Gua$^{npe^6}_{ibu}$	$(MeO)_2Tr$
78	Gua$^{npe^6}_{npeoc}$	$(MeO)_2Tr$
79	Thy$^{me^4}$	$(MeO)_2Tr$
80	Thy$^{et^4}$	$(MeO)_2Tr$
81	Thy$^{ipr^4}$	$(MeO)_2Tr$
82	Thy$^{npe^4}$	$(MeO)_2Tr$

npe = 2-(4-nitrophenyl)ethyl
npeoc = [2-(4-nitrophenyl)ethoxy]carbonyl
ibu = isobutyryl = 2-methyl-1-oxopropyl
ipr = Me_2CH
nbu = $Me(CH_2)_3$
MeOTr = (4-methoxyphenyl)diphenylmethyl
$(MeO)_2Tr$ = bis(4-methoxyphenyl)phenylmethyl

pylphosphorodiamidite (= bis(diisopropylamino)[2-(4-nitrophenyl)ethoxy]phosphane; **51**) [39], respectively, the 3′-[2-(4-nitrophenyl)ethyl diisopropylphosphoramidites] **67**–**82** by common procedures. Workup and purification were achieved by silica gel column chromatography using efficiently AcOEt/Et_3N or toluene/AcOEt in the elution process.

3. Physical Data. – The purity and structures of the newly synthesized compounds were established by chromatographical means and by UV, ^{1}H-NMR, and ^{31}P-NMR spectra, as well as by C,H,N analyses.

Experimental Part

General. TLC: precoated silica-gel thin-layer sheets *60 F 254* from *Merck*. Prep. column chromatography (CC): silica gel (*Merck 60*, 63–200 μm). M.p.: *Gallenkamp* melting-point apparatus; no correction. UV/VIS: *Perkin-Elmer Lambda 15*; λ_{max} in nm (log ε). IR: *Perkin-Elmer FTIR-1600*; $\tilde{\nu}$ in cm^{-1}. ^{1}H-NMR: *Bruker WM-250*; δ in ppm rel. to $SiMe_4$. ^{31}P-NMR: *Jeol JM GX-400*; δ in ppm rel. to 85% H_3PO_4 soln.

1. *2′-Deoxy-*N^2*-[2-(4-nitrophenyl)ethoxycarbonyl]guanosine* (**5**). The 2′-deoxyguanosine (**2**) (0.267 g, 1 mmol) was co-evaporated several times with dry pyridine (3 × 2 ml), then suspended in pyridine (20 ml), and treated with Me_3SiCl (0.543 g, 0.64 ml, 5 mmol) by stirring at r.t. for 30 min. After cooling in an ice-bath, 2-(4-nitrophenyl)ethyl carbonochloridite [12] (0.459 g, 2 mmol) in $CHCl_3$ (4 ml) was added dropwise within 30 min and stirred at 0° for 1 h and at r.t. for 2 days. MeOH (10 ml) was then added to the clear soln. and stirred at r.t. for 5 h. The solvent was evaporated, the residue treated overnight in a 10% $NaHCO_3$ soln., and the resulting colorless precipitate washed with Et_2O and dried at 50° *in vacuo:* 0.23 g (50%) of **5**. TLC ($CHCl_3$/MeOH 4:1): R_f 0.36. M.p. 185–190°. UV (MeOH): 272 (sh, 4.34), 258 (4.41), 251 (sh, 4.37). ^{1}H-NMR ((D$_6$)DMSO): 11.46, 11.32 (2 *s*, NH); 8.21 (*s*, H–C(8)); 8.17 (*d*, 2 H *o* to NO_2); 7.63 (*d*, 2 H *m* to NO_2); 6.21 (*t*, H–C(1′)); 5.32 (*d*, OH–C(3′)); 4.95 (*t*, OH–C(5′)); 4.48 (*t*, OC*H*$_2$CH$_2$ (npeoc)); 4.36 (*m,* H–C(3′)); 3.82 (*m*, H–C(4′)); 3.59–3.45 (*m*, 2 H–C(5′)); 3.14 (*t*, OCH$_2$C*H*$_2$ (npeoc)); 2.99 (*m*, 1 H–C(2′)); 2.55 (*m*, 1 H–C(2′)). Anal. calc. for $C_{18}H_{20}N_6O_8$ (460.4): C 49.57, H 4.38, N 18.25; found: C 49.21, H 4.19, N 18.20.

2. *2′-Deoxy-*N^2*-[2-(4-nitrophenyl)ethoxycarbonyl]-*O^6*-[2-(4-nitrophenyl)ethyl]guanosine* (**7**). A soln. of 3′,5′-di-*O*-acetyl-2′-deoxyguanosine (0.69 g, 2 mmol) [15], triphenylphosphine (0.84 g, 3.2 mmol), and 2-(4-nitrophenyl)ethanol [40] (0.50 g, 3 mmol) in dry dioxane (40 ml) was stirred at r.t. for a few min and then treated with diethyl diazenedicarboxylate (0.558 g, 3.2 mmol) with stirring for 1 h. The clear soln. was evaporated and co-evaporated with dry pyridine (40 ml). The residue was taken up in pyridine (10 ml), cooled in an ice-bath, and then treated dropwise with 2-(4-nitrophenyl)ethyl carbonochloridate [12] (1.38 g, 6 mmol) in dry $CHCl_3$ (10 ml). After 1 h, stirring was continued at r.t. for 3 h. Then the mixture was diluted with H_2O (100 ml) and extracted with $CHCl_3$ (4 × 50 ml), the org. phase dried (Na_2SO_4), evaporated, and co-evaporated with toluene, and the residue purified by CC (silica gel, CH_2Cl_2, then $CHCl_3$). The main fraction was evaporated and the resulting residue treated with dioxane (25 ml) and 25% NH_3 soln. (25 ml) by keeping the mixture in the icebox for 20 h. After evaporation, the residue was recrystallized from MeOH (80 ml): 0.81 g (66%) of **7**. TLC ($CHCl_3$/MeOH 9:1): R_f 0.53. M.p. 179–182°. UV (MeOH): 269 (4.54), 216 (4.63). ^{1}H-NMR ((D$_6$)DMSO): 10.33 (*s*, NH); 8.40 (*s*, H–C(8)); 8.17 (*d*, 2 H *o* to NO_2); 7.64 (*d*, 2 H *m* to NO_2); 6.30 (*t*, H–C(1′)); 5.32 (*d*, OH–C(3′)); 4.89 (*t*, OH–C(5′)); 4.73 (*t*, OC*H*$_2$CH$_2$, npe); 4.41 (*m,* H–C(3′)); 4.37 (*t*, OC*H*$_2$CH$_2$ (npeoc)); 3.83 (*m*, H–C(4′)); 3.67–3.42 (*m*, 2 H–C(5′)); 3.30 (*t*, OCH$_2$C*H*$_2$ (npeoc)); 3.11 (*t*, OCH$_2$C*H*$_2$ (npe)); 2.71 (*m*, 1 H–C(2′)); 2.25 (*m*, 1 H–C(2′)). Anal. calc. for $C_{27}H_{27}N_7O_{10} \cdot 0.5H_2O$ (618.5): C 52.42, H 4.56, N 15.85; found: C 52.32, H 4.67, N 15.63.

3. *2′-Deoxy-*N^2*-isobutyryl-*O^6*-methylguanosine* (= *2′-Deoxy-*O^6*-methyl-*N^2*-(2-methyl-1-oxopropyl)guanosine*; **9**) [21]. A soln. of 2′-deoxy-*N*^{2},3′-*O*,5′-*O*-triisobutyrylguanosine (5 g, 10.47 mmol) [20], triphenylphosphine (3.44 g, 13.09 mmol), and MeOH (0.63 ml, 15.71 mmol) in dry dioxane (40 ml) was stirred at r.t. for a few min and then treated with diethyl diazenedicarboxylate (2.2 g, 2 ml, 13.09 mmol) and stirred further for 24 h. The clear soln. was evaporated and the residue dissolved in CH_2Cl_2 and purified by CC (silica gel, (48 × 3 cm), Et_2O/petroleum ether (3:1, Et_2O). The product fractions were collected and then evaporated to give **8.** To a soln. of this solid in dry MeOH (40 ml), 0.5M NaOMe/MeOH (8 ml) was added and stirred at r.t. for 1 h. The mixture was neutralized with 1M AcOH (5 ml) and evaporated. The residue was dissolved in MeOH, and the soln. mixed with silica gel (20 g) and evaporated. The dried powder was applied to CC (silica gel (10 × 3.5 cm),

$CHCl_3$, then $CHCl_3$/MeOH 19:1): 1.3 g (35%) of **9**. TLC ($CHCl_3$/MeOH 9:1): R_f 0.38. M.p. 183–184°. UV (MeOH): 268 (4.21), 218 (4.31). ^{1}H-NMR ((D_6)DMSO): 9.52 (*s*, NH); 7.57 (*s*, H–C(8)); 5.48 (*t*, H–C(1′)); 4.46 (*d*, OH–C(3′)); 4.07 (*t*, OH–C(5′)); 3.56 (*m*, H–C(3′)); 3.21 (*s*, MeO–C(6)); 3.09 (*m*, H–C(4′)); 2.75 (*m*, 2 H–C(5′)); 2.03 (*m*, 1 H–C(2′)); 1.83 (*m*, 1 H–C(2′)); 1.46–1.37 (*m*, Me_2CHCO); 0.24–0.22 (*d*, Me_2CHCO). Anal. calc. for $C_{15}H_{21}N_5O_5$ (351.4): C 51.27, H 6.02, N 19.93; found: C 51.30, H 6.22, N 19.66.

4. *2′-Deoxy-*O^6*-ethyl-*N^2*,3′-*O*,5′-*O*-triisobutyrylguanosine* (= *2′-Deoxy-*O^6*-ethyl-*N^2*-(2-methyl-1-oxopropyl)-guanosine 3′,5′-Bis(2-methylpropanoate)*; **10**) [21]. A soln. of 2′-deoxy-*N*^{2}, 3′-*O*,5′-*O*-triisobutyrylguanosine (5 g, 10.47 mmol) [20], triphenylphosphine (3.44 g, 13.09 mmol), and EtOH (0.9 ml, 15.7 mmol) in dry dioxane (40 ml) was stirred at r.t. for a few min and then treated with diethyl diazenedicarboxylate (2.2 g, 2.0 ml, 13 mmol) by stirring for 24 h. The clear soln. was evaporated and purified by CC (silica gel, Et_2O): 3.78 g (71%) of **10**. Amorphous solid. TLC (Et_2O): R_f 0.25. UV (MeOH): 267 (4.23), 218 (4.35). ^{1}H-NMR ($CDCl_3$): 7.95 (*s*, NH); 7.93 (*s*, H–C(8)); 6.36 (*t*, H–C(1′)); 5.38 (*m*, H–C(3′)); 4.60 (*t*, MeC*H*$_2$O–C(6)); 4.49–4.28 (*m*, H–C(4′), 2 H–C(5′)); 3.00–2.90 (*m*, 2 Me_2CHCO); 2.62–2.49 (*m*, 1 Me_2CHCO, 2 H–C(2′)); 1.50 (*t*, *Me*CH$_2$O–C(6)); 1.27–1.11 (*m*, 3 Me_2CHCO). Anal. calc. for $C_{24}H_{35}N_5O_7$ (505.6): C 57.01, H 6.98, N 13.85; found: C 56.76, H 7.23, N 13.51.

5. *2′-Deoxy-*O^6*-ethyl-*N^2*-isobutyrylguanosine* (= *2′-Deoxy-*O^6*-ethyl-*N^2*-(2-methyl-1-oxopropyl)guanosine*; **11**) [21]. To a soln. of **10** (8.09 g, 16 mmol) in dry EtOH (40 ml), 0.5M NaOEt/EtOH (8 ml) was added. The mixture was stirred at r.t. for 1 h, then neutralized with 1M AcOH (5 ml), and evaporated . The residue was dissolved in EtOH, mixed with silica gel (20 g) and evaporated. The dried powder was applied to CC (silica gel 10 × 3.5 cm), $CHCl_3$, then $CHCl_3$/MeOH 19:1): 5.6 g (95%) of **11**. TLC ($CHCl_3$/MeOH 9:1): R_f 0.45. M.p. 160–161°. UV (MeOH): 269 (4.24), 219 (4.35). ^{1}H-NMR ((D_6)DMSO): 9.48 (*s*, NH); 7.57 (*s*, H–C(8)); 5.48 (*t*, H–C(1′)); 4.48 (*d*, OH–C(3′)); 4.05 (*t*, OH–C(5′)); 3.76 (*q*, MeC*H*$_2$O–C(6)); 3.58 (*m*, 2 H–C(3′)); 3.00 (*m*, H–C(4′)); 2.73 (*m*, 2 H–C(5′)); 2.03 (*m*, 1 H–C(2′)); 1.83 (*m*, 1 H–C(2′)); 1.44 (*m*, Me_2CHCO); 0.58 (*t*, *Me*CH$_2$O–C(6)); 0.25–0.23 (*d*, Me_2CHCO). Anal. calc. for $C_{16}H_{23}N_5O_5$ (365.4): C 52.59, H 6.35, N 19.61; found: C 52.30, H 6.45, N 19.00.

6. *2′-Deoxy-*N^2*,3′-*O*,5′-*O*-triisobutyryl-*O^6*-isopropylguanosine* (= *2′-Deoxy-*O^6*-(2-methylethyl)-*N^2*-(2-methyl-1-oxopropyl)guanosine 3′,5′-Bis(2-methylpropanoate)*; **12**) [21]. As described for **10**, with 2′-deoxy-*N*^{2},3′-*O*,5′-*O*-triisobutyrylguanosine (5 g, 10.47 mmol) [20], triphenylphosphine (3.44 g, 13.1 mmol), ^{i}PrOH (0.95 g, 1.2 ml, 16 mmol), and dioxane (40 ml): 4.40 g (81%) of **12**. UV (MeOH): 267 (4.23), 218 (4.37). ^{1}H-NMR ($CDCl_3$): 7.94 (*s*, NH); 7.91 (*s*, H–C(8)); 6.36 (*t*, H–C(1′)); 5.57 (*m*, Me_2CHO–C(6)); 5.38 (*m*, H–C(3′)); 4.48–4.35 (*m*, H–C(4′)); 4.29 (*m*, H–C(5′)); 2.99–2.89 (*m*, Me_2CHCOO, 1 H–C(2′)); 2.56 (*m*, Me_2CHCON, 1 H–C(2′)); 1.43 (*d*, Me_2CHO–C(6)); 1.26–1.10 (*m*, Me_2CHCO). Anal. calc. for $C_{23}H_{37}N_5O_7$ (519.6): C 57.78, H 7.18, N 13.47; found: C 57.58, H 7.33, N 13.23.

7. *2′-Deoxy-*N^2*-isobutyryl-*O^6*-isopropylguanosine* (= *2′-Deoxy-*O^6*-(2-methylethyl)-*N^2*-(2-methyl-1-oxopropyl)guanosine*; **13**) [21]. As described for **11**, with **12** (8.3 g, 16 mmol): 5.84 g (96%) of **13**. TLC ($CHCl_3$/MeOH 95:5): R_f 0.29 . UV (MeOH): 268 (4.23), 218 (4.35). ^{1}H-NMR ((D_6)DMSO): 10.63 (*s*, NH); 7.57 (*s*, H–C(8)); 5.47 (‘*t*’, H–C(1′)); 4.73 (*m*, Me_2CHO–C(6)); 4.46 (*d*, OH–C(3′)); 4.06 (*t*, OH–C(5′)); 3.57 (*m*, H–C(3′)); 3.01 (*m*, H–C(4′)); 2.70 (*m*, 2 H–C(5′)); 2.02, 1.99 (2 *m*, 2 H–C(2′)); 1.54 (*m*, Me_2CHCO); 0.54 (*d*, Me_2CHO–C(6)); 0.26–0.22 (*d*, Me_2CHCO). Anal. calc. for $C_{17}H_{25}N_5O_5$ (379.4): C 53.81, H 6.64, N 18.45; found: C 53.01, H 6.90, N 18.03.

8. *O^6-Butyl-2′-deoxy-*N^2*-isobutyrylguanosine* (= *O^6-Butyl-2′-deoxy-*N^2*-(2-methyl-1-oxopropyl)guanosine*; **15**). As described for **9**, with 2′-deoxy-*N*^{2},3′-*O*,5′-*O*-triisobutyrylguanosine (5 g, 10.47 mmol) [20] and butan-1-ol (1.4 ml, 15.71 mmol) *via* the intermediate **14**: 2.9 g (71%) of **15**. TLC ($CHCl_3$/MeOH 95:5): R_f 0.29. UV (MeOH): 268 (4.30), 218 (4.43). ^{1}H-NMR ($CDCl_3$): 7.86 (*s*, NH, H–C(8)); 6.28 (*t*, H–C(1′)); 4.84 (*d*, OH–C(3′)); 4.65 (*t*, OH–C(5′)); 4.48 (*t*, $MeCH_2CH_2CH_2O$); 4.10 (*m*, H–C(3′)); 3.86 (*m*, 2 H–C(5′)); 3.11 (*m*, H–C(4′)); 2.89 (*m*, Me_2CHCO, 1 H–C(2′)); 2.35 (*m*, H–C(2′)); 1.77 (*m*, $MeCH_2CH_2CH_2O$); 1.45 (*m*, $MeCH_2CH_2CH_2O$); 1.22 (*d*, Me_2CHCO); 0.90 (*t*, *Me*$CH_2CH_2CH_2O$). Anal. calc. for $C_{18}H_{27}N_5O_5$ (393.3): C 54.95, H 6.92, N 17.80; found: C 54.01, H 6.95, N 17.52.

9. *3′,5′-Di-*O*-acetyl-2′-deoxy-*O^6*-methylguanosine* (**16**) *and 3′,5′-Di-*O*-acetyl-2′-deoxy-*N^1*-methylguanosine* (**17**). A suspension of 3′,5′-di-*O*-acetyl-2′-deoxyguanosine [15] (20 g, 57 mmol), triphenylphosphine (28.3 g, 107 mmol), and dry MeOH (4.4 ml, 3.3 g, 110 mmol) in dry dioxane (750 ml) was stirred at r.t. for a few min. Then diethyl diazenedicarboxylate (17.5 ml, 18.6 g, 3.2 mmol) was added and the mixture stirred further for 24 h. The clear soln. was evaporated and the residue purified by FC (Et_2O, then AcOEt and AcOEt/MeOH 9:1): 5.88 g (25%) of **16**, followed by 9.0 g (38%) of **17**, after drying in a vacuum desiccator.

Data of **16**: TLC ($CHCl_3$/MeOH 19:1): R_f 0.50. UV (MeOH): 280 (3.99), 248 (4.03), 209 (4.38). ^{1}H-NMR ($CDCl_3$): 7.72 (*s*, H–C(8); 6.26 (*dd*, H–C(1′)); 5.42 (*m*, H–C(3′)); 4.88 (*s*, NH_2); 4.46–4.31 (*m*, H–C(4′),

2 H–C(5′)); 4.05 (*s*, MeO–C(6)); 2.97 (*m*, 1 H–C(2′)); 2.55 (*m*, 1 H–C(2′)); 2.22, 2.07 (2 *s*, 2 Ac). Anal. calc. for $C_{15}H_{19}N_5O_6$ (365.4): C 49.32, H 5.25, N 19.17; found: C 48.77, H 5.52, N 18.41.

Data of **17**: TLC ($CHCl_3$/MeOH 19:1): R_f 0.16. UV (MeOH): 272 (sh, 4.02), 257 (4.15), 203 (4.25). ^{1}H-NMR ($CDCl_3$): 7.62 (*s*, H–C(8)); 6.20 (*dd*, H–C(1′)); 5.41 (*m*, H–C(3′)); 5.03 (*s*, NH_2); 4.53–4.29 (*m*, H–C(4′), 2 H–C(5′)); 3.50 (*s*, Me–N(1)); 2.95 (*m*, 1 H–C(2′)); 2.51 (*m*, 1 H–C(2′)); 2.11, 2.08 (2 *s*, 2 Ac). Anal. calc. for $C_{15}H_{19}N_5O_6 \cdot 0.2\ CHCl_3$ (389.2): C 46.91, H 4.97, N 17.99; found: C 47.00, H 5.24, N 17.95.

10. *3′,5′-Di*-O-*acetyl-2′-deoxy*-O^6-*methyl*-N^2-*[2-(4-nitrophenyl)ethoxycarbonyl]guanosine* (**18**). To a soln. of **16** (5.7 g, 15.6 mmol) in dry pyridine (70 ml) cooled to 0° in an ice-bath, 2-(4-nitrophenyl)ethyl carbonochloridate [12] (5.12 g, 22.3 mmol) was added dropwise slowly. After stirring for 1 h and another 20 h at r.t., the mixture was evaporated and co-evaporated with toluene (50 ml), and the residue was purified by FC (silica gel, $CHCl_3$/MeOH 98:2): 7.59 g (87%) of **18**. Foam. TLC ($CHCl_3$/MeOH 19:1): R_f 0.61. UV (MeOH): 267 (4.23), 256 (sh, 4.17), 216 (4.39). ^{1}H-NMR ($CDCl_3$): 8.16 (*d*, 2 H *o* to NO_2); 7.91 (*s*, H–C(8)); 7.50 (*s*, NH); 7.41 (*d*, 2 H *m* to NO_2); 6.34 (*t*, H–C(1′)); 5.48 (*m*, H–C(3′)); 4.49–4.28 (*m*, 5 H, OCH_2CH$_2$ (npeoc), H–C(4′), 2 H–C(5′)); 4.09 (*s*, MeO–C(6)); 3.11 (*t*, OCH$_2$CH_2 (npeoc)); 3.07 (*m*, 1 H–C(2′)); 2.58 (*m*, 1 H–C(2′)); 2.10, 2.04 (2 *s*, Ac). Anal. calc. for $C_{24}H_{26}N_6O_{10} \cdot 1.25\ H_2O$ (581.0): C 49.61, H 4.94, N 14.46; found: C 49.63, H 4.43, N 14.30.

11. *2′-Deoxy*-O^6-*methyl*-N^2-*[2-(4-nitrophenyl)ethoxycarbonyl]guanosine* (**19**). To a soln. of **18** (7 g, 12.5 mmol) in dioxane (25 ml), 25% NH_3 soln. (25 ml) was added and stirred at 5° for 20 h. The solvent was evaporated and co-evaporated with MeOH/toluene 1:1 and then the product crystallized from AcOEt/MeOH (50 ml): 5.23 g (88%) of **19**. TLC ($CHCl_3$/MeOH 19:1): R_f 0.13. UV (MeOH): 267 (4.29), 256 (sh, 4.23), 216 (4.43). ^{1}H-NMR ((D$_6$)DMSO): 10.29 (*s*, NH); 8.40 (*s*, H–C(8)); 8.17 (*d*, 1 H *o* to NO_2); 7.62 (*d*, 1 H *m* to NO_2); 6.31 (*dd*, H–C(1′)); 5.30 (*d*, OH–C(3′)); 4.88 (*d*, OH–C(5′)); 4.40 (*m*, H–C(3′)); 4.30 (*t*, OCH_2CH$_2$ (npeoc)); 4.03 (*s*, MeO–C(6)); 3.85 (*m*, H–C(4′)); 3.63–3.38 (*m*, 2 H–C(5′)); 3.09 (*t*, OCH$_2$CH_2 (npeoc)); 2.77 (*m*, 1 H–C(2′)); 2.30 (*m*, 1 H–C(2′)). Anal. calc. for $C_{20}H_{22}N_6O_8 \cdot 0.7\ H_2O$ (487.4): C 49.32, H 4.84, N 17.26; found: C 49.33, H 4.49, N 17.24.

12. O^4-*Butylthymidine* (**23**). a) To a soln. of NaOBu (prepared from Na (0.08 g, 3.0 mmol) in BuOH (20 ml), 3′,5′-di-*O*-acetyl-*O*^{4}-(1*H*-triazol-1-yl)thymidine [23] (1.13 g, 3.0 mmol) was added and stirred at r.t. for 1 h. MeOH (10 ml) was then added, the mixture neutralized with 1M HCl, evaporated, and co-evaporated with MeOH, and the residue purified by CC (silica gel, $CHCl_3$/MeOH 20:1): 0.67 g (75%) of **23**. Colorless foam.

b) A mixture of 3′,5′-di-*O*-acetyl-*O*^{4}-(1*H*-triazol-1-yl)thymidine (2.83 g, 7.5 mmol), 4-(dimethylamino)-pyridine (2.75 g, 22.5 mmol), BuOH (7.5 ml, 82 mmol), and MeCN (40 ml) was heated under reflux for 27 h. After evaporation, the residue was purified by CC (silica gel, AcOEt). The product fractions were evaporated, and the residue was taken up in MeOH (10 ml) and treated with 25% NH_3 soln. (5 ml). After stirring at r.t. for 15 h, the solvent was evaporated and co-evaporated with MeOH. Purification was achieved by CC ($CHCl_3$/MeOH 100:5): 1.75g (78%) of **23**. TLC ($CHCl_3$/MeOH 9:1): R_f 0.68. UV (MeOH): 283 (3.81), 220 (sh, 4.01), 206 (4.25). ^{1}H-NMR ((D$_6$)DMSO): 8.00 (*s*, H–C(6)); 6.13 (*t*, H–C(1′)); 5.23 (*d*, OH–C(3′)); 5.06 (*t*, OH–C(5′)); 4.24 (*m*, H–C(3′), MeCH$_2$CH$_2$CH_2O); 3.79 (*m*, H–C(4′)); 3.59 (*m*, 2 H–C(5′)); 2.18 (*m*, 1 H–C(2′)); 1.99 (*m*, 1 H–C(2′)); 1.86 (*s*, Me–C(5)); 1.65 (*m*, MeCH$_2$CH_2CH$_2$O); 1.37 (*m*, MeCH_2CH$_2$CH$_2$O); 0.90 (*t*, *Me*CH$_2$CH$_2$CH$_2$O). Anal. calc. for $C_{14}H_{22}N_2O_5$ (298.4): C 56.36, H 7.43, N 9.39; found: C 56.26, H 7.31, N 9.47.

13. *2′-Deoxy-5′*-O-*(4-monomethoxytrityl)*-N^2-*[2-(4-nitrophenyl)ethoxycarbonyl]*-O^6-*[2-(4-nitrophenyl)-ethyl)]guanosine* (**28**). To a soln. of dried **7** (4.27 g, 7 mmol) in dry pyridine (35 ml), 4-monomethoxytrityl chloride (2.81 g, 9.1 mmol) was added and the mixture stirred at r.t. for 16 h. After completion of the reaction, MeOH (10 ml) was added and the mixture concentrated to *ca.* 3/4 of the volume, diluted with $CHCl_3$ (100 ml), and washed with H_2O (2 × 20 ml). The org. layer was dried (Na_2SO_4), evaporated, and co-evaporated with toluene. Purification by CC (silica gel, $CHCl_3$/MeOH 100:1) gave 5.55 g (90%) of **28**. Colorless foam. TLC ($CHCl_3$/MeOH 95:5): R_f 0.49. UV (MeOH): 269 (4.55), 236 (4.37). ^{1}H-NMR ($CDCl_3$): 8.12–8.01 (2 *d*, 4 H *o* to NO_2); 7.96 (*s*, H–C(8)); 7.51 (*s*, NH); 7.48–7.10 (*m*, 12 arom. H, 4 H *m* to NO_2); 6.73 (*d*, 2 H *o* to MeO); 6.56 (*t*, H–C(1′)); 4.73 (*m*, H–C(3′), OCH_2CH$_2$ (npeoc)); 4.34 (*t*, OCH_2CH$_2$ (npe)); 4.20 (*m*, H–C(4′)); 3.72 (*s*, MeO); 3.57 (*m*, OH–C(3′)); 3.35 (*m*, 1 H–C(5′)); 3.25 (*t*, OCH$_2$CH_2 (npeoc)); 3.02 (*t*, OCH$_2$CH_2 (npe)); 2.70 (*m*, 1 H–C(2′)); 2.56 (*m*, 1 H–C(2′)). Anal. calc. for $C_{47}H_{43}N_7O_{11}$ (881.9): C 64.01, H 4.91, N 11.12; found: C 63.89, H 5.05, N 10.92.

14. *General Procedure A (G.P. A): Synthesis of 5′*-O-*(4,4′-Dimethoxytrityl)- and Base-Protected 2′-Deoxyribonucleosides.* To a soln. of dried compounds **3**–**7**, **9**, **11**, **13**, **15**, **19**, and **20**–**24** (45 mmol) in pyridine (200 ml) 4,4′-dimethoxytrityl chloride (18.3 g, 53 mmol) was added and stirred at r.t. for 2 to 20 h (TLC control). After completion of the reaction, MeOH (10 ml) was added and the mixture evaporated to *ca.* 1/4 of the volume. The mixture was diluted with CH_2Cl_2 (200 ml) and washed with H_2O (2 × 100 ml). The org. layer was dried

(Na_2SO_4), evaporated, and co-evaporated with toluene. Purification by CC or FC (silica gel) using the appropriate solvents gave the required compound as a colorless foam.

15. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*N^6*-[2-(4-nitrophenyl)ethoxycarbonyl]adenosine* (**33**). From **3** [12] by *G.P. A*. FC ($CHCl_3$/MeOH 100 : 4). Yield 84%. TLC ($CHCl_3$/MeOH 95 : 5): R_f 0.34. UV (MeOH): 276 (sh, 4.41), 268 (4.47), 236 (4.43). ^{1}H-NMR ($CDCl_3$): 8.66 (*s*, H–C(8)); 8.36 (*br. s*, NH); 8.16 (*d*, 1 H *o* to NO_2); 8.11 (*s*, H–C(2)); 7.44–7.15 (*m*, 9 arom. H); 6.80 (*d*, 2 H *o* to MeO); 6.44 (*m*, H–C(1′)); 4.71 (*m*, H–C(3′)); 4.53 (*t*, OCH_2CH_2 (npeoc)); 4.24 (*m*, H–C(4′)); 3.77 (*s*, 2 MeO); 3.52–3.40 (*m*, 2 H–C(5′)); 3.14 (*t*, OCH_2CH_2 (npeoc)); 2.91–2.51 (*m*, 2 H–C(2′)). Anal. calc. for $C_{40}H_{38}N_6O_9 \cdot 0.5\ H_2O$ (755.8): C 63.57, H 5.20, N 11.12; found: C 63.62, H 5.21, N 10.95.

16. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*N^2*-[2-(4-nitrophenyl)ethoxycarbonyl]guanosine* (**35**). From **5** by *G.P. A*. Purification by CC ($CHCl_3$/Et_3N 100 : 0.2 → $CHCl_3$/MeOH/Et_3N 90 : 10 : 0.2). Yield 79%. TLC ($CHCl_3$/MeOH 95 : 5): R_f 0.40. UV (MeOH): 272 (sh, 4.38), 258 (4.42), 249 (4.42), 237 (4.46). ^{1}H-NMR ($CDCl_3$): 11.27 (*s*, NH); 9.76 (*s*, NH); 8.00 (*d*, 2 H *o* to NO_2); 7.75 (*s*, H–C(8)); 7.32–7.03 (*m*, arom. H); 6.67 (*d*, 2 H *o* to MeO); 6.15 (*t*, H–C(1′)); 4.80 (*m*, H–C(3′)); 4.44 (*m*, OH–C(3′), OCH_2CH_2 (npeoc)); 4.15 (*m*, H–C(4′)); 3.65 (*s*, MeO); 3.30 (*m*, 2 H–C(5′)); 3.01 (*m*, OCH_2CH_2 (npeoc)); 2.60–2.46 (*m*, 2 H–C(2′)). Anal. calc. for $C_{40}H_{38}N_6O_{10}$ (762.8): C 62.99, H 5.02, N 11.02; found: C 62.43, H 5.14, N 10.93.

17. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*N^2*-[2-(4-nitrophenyl)ethoxycarbonyl]-*O^6*-[2-(4-nitrophenyl)ethyl]-guanosine* (**37**). From **7** by *G.P. A*. Purification by CC ($CHCl_3$/Et_3N 100 : 0.2 → $CHCl_3$/MeOH/Et_3N 99 : 1 : 0.2). Yield 85%. TLC ($CHCl_3$/MeOH 95 : 5): R_f 0.43. UV (MeOH): 268 (4.55), 235 (4.47). ^{1}H-NMR ($CDCl_3$): 8.15–8.11 (*m*, NH, 2 H *o* to NO_2); 7.96 (*s*, H–C(8)); 7.50–7.13 (*m*, 9 arom. H); 6.77 (*d*, 4 H *o* to MeO); 6.48 (*m*, H–C(1′)); 4.81–4.74 (*t*, *m*, H–C(3′), OCH_2CH_2 (npe)); 4.45 (*t*, 2 OCH_2CH_2 (npeoc)); 4.16 (*m*, H–C(4′)); 3.75 (*s*, 2 MeO); 3.56–3.25 (*m*, 2 H–C(5′), OCH_2CH_2 (npe)); 3.09–3.03 (*t*, *br. s*, OH–C(3′), OCH_2CH_2 (npeoc)); 2.82–2.70 (*m*, 1 H–C(2′)); 2.55 (*m*, 1 H–C(2′)). Anal. calc. for $C_{48}H_{45}N_7O_{12}$ (911.9): C 63.22, H 4.97, N 10.75; found: C 63.08, H 5.14, N 10.67.

18. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*N^2*-isobutyryl-*O^6*-methylguanosine* (**38**). From **9** by *G.P. A*. FC ($CHCl_3$/MeOH 19 : 1). Yield 93%. TLC ($CHCl_3$/MeOH 19 : 1): R_f 0.52. UV (MeOH): 280 (sh, 4.32), 269 (4.38), 234 (4.49), 219 (sh, 4.76), 206 (4.94). ^{1}H-NMR ($CDCl_3$): 8.26 (*s*, NH); 8.11 (*s*, H–C(8); 7.50–7.24 (*m*, 9 arom. H); 6.85 (*d*, 4 H *o* to MeO); 6.78 (*t*, H–C(1′)); 4.81 (*m*, H–C(3′)); 4.39 (*s*, OH–C(3′)); 4.37 (*d*, H–C(4′)); 4.18 (*s*, MeO–C(6)); 3.83 (*s*, 2 MeO); 3.51–3.40 (*m*, 2 H–C(5′)); 2.77–2.72 (*m*, Me_2CHCO, 2 H–C(2′)); 1.29–1.25 (*d*, Me_2CHCO). Anal. calc. for $C_{36}H_{39}N_5O_7$ (653.7): C 66.14, H 6.01, N 11.01; found: C 65.78, H 6.08, N 10.81.

19. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*N^2*-isobutyryl-*O^6*-ethylguanosine* (**39**). From **11** by *G.P. A*. FC ($CHCl_3$/MeOH 19 : 1). Yield 95%. TLC ($CHCl_3$/MeOH 19 : 1): R_f 0.54. UV (MeOH): 280 (sh, 4.10), 269 (4.26), 234 (sh, 4.37), 216 (sh, 4.57), 203 (4.80). ^{1}H-NMR ($CDCl_3$): 8.02 (*s*, H–C(8)); 8.01 (*s*, NH); 7.44–7.19 (*m*, 9 arom. H); 6.81–6.78 (*d*, 4 H *o* to MeO); 6.65 (*t*, H–C(1′)); 4.75 (*m*, H–C(3′)); 4.64 (*q*, $MeCH_2O$–C(6)); 4.24 (*d*, H–C(4′)); 3.78 (*s*, 2 MeO); 3.46–3.33 (*m*, 2 H–C(5′)); 3.00 (*br. s*, OH–C(3′)); 2.75–2.64 (*m*, Me_2CHCO, 2 H–C(2′)); 1.51 (*t*, $MeCH_2O$–C(6)); 1.26 (*m*, Me_2CHCO). Anal. calc. for $C_{37}H_{41}N_5O_7$ (667.7): C 66.55, H 6.18, N 10.48; found: C 66.14, H 6.30, N 10.30.

20. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*N^2*-isobutyryl-*O^6*-isopropylguanosine* (**40**). From **13** by *G.P. A*. FC ($CHCl_3$/MeOH 19 : 1). Yield 95%. TLC ($CHCl_3$/MeOH 19 : 1): R_f 0.56. UV (MeOH): 280 (sh, 4.07), 265 (4.27), 234 (sh, 4.36), 203 (4.83). ^{1}H-NMR ($CDCl_3$): 7.95 (*s*, H–C(8)); 7.76 (*s*, NH); 7.41–7.18 (*m*, 9 arom. H); 6.78 (*d*, 4 H *o* to MeO); 6.49 (*t*, H–C(1′)); 5.57 (*q*, Me_2CHO–C(6)); 4.72 (*m*, H–C(3′)); 4.14 (*d*, H–C(4′)); 3.77 (*s*, 2 MeO); 3.46–3.32 (*m*, 2 H–C(5′)); 3.00 (*br. s*, OH–C(3′)); 2.81–2.73 (*m*, Me_2CHCO, 1 H–C(2′)); 2.59 (*m*, 1 H–C(2′)); 1.46 (*d*, Me_2CHO–C(6)); 1.25–1.19 (2 *d*, Me_2CHCO). Anal. calc. for $C_{38}H_{43}N_5O_7 \cdot 0.5\ H_2O$ (690.8): C 66.07, H 6.42, N 10.13; found: C 66.16, H 6.50, N 9.88.

21. *O^6-Butyl-2′-deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*N^2*-isobutyrylguanosine* (**41**). From **15** by *G.P. A*. FC ($CHCl_3$/MeOH 98 : 2). Yield 85%. TLC ($CHCl_3$/MeOH 95 : 5): R_f 0.82. UV (MeOH): 280 (sh, 4.14), 269 (4.30), 234 (sh, 4.41). ^{1}H-NMR ($CDCl_3$): 8.02 (2 *s*, H–C(8), NH); 7.41–7.20 (*m*, 9 arom. H); 6.77 (*d*, 4 H *o* to MeO); 6.63 (*t*, H–C(1′)); 4.73 (*m*, H–C(3′)); 4.53 (*t*, $MeCH_2CH_2CH_2O$); 4.23 (*d*, H–C(4′)); 3.75 (*s*, 6 MeO); 3.40–3.32 (*m*, 2 H–C(5′)); 2.91 (*br. s*, OH–C(3′)); 2.69–2.60 (2 *m*, Me_2CHCO, 2 H–C(2′)); 1.85 (*m*, $MeCH_2CH_2CH_2O$); 1.52 (*m*, $MeCH_2CH_2CH_2O$); 1.21–1.17 (2 *d*, Me_2CHCO); 0.96 (*t*, $MeCH_2CH_2CH_2O$). Anal. calc. for $C_{39}H_{45}N_5O_7$ (695.8): C 67.32, H 6.52, N 10.07; found: C 67.30, H 6.56, N 10.10.

22. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*O^6*-methyl-*N^2*-[2-(4-nitrophenyl)ethoxycarbonyl]guanosine* (**42**). From **19** by *G.P. A*. CC ($CHCl_3$/MeOH/Et_3N 495 : 4 : 1). Yield 89%. TLC ($CHCl_3$/MeOH 19 : 1): R_f 0.52. UV (MeOH): 268 (4.13), 236 (4.15), 204 (4.58). ^{1}H-NMR ($CDCl_3$): 8.17 (*d*, 2 H *o* to NO_2); 7.93 (*s*, H–C(8)); 7.41–7.17 (*m*, 12 arom. H, NH); 6.77 (*m*, 4 H *o* to MeO); 6.42 (*dd*, H–C(1′)); 4.81 (*m*, H–C(3′)); 4.43 (*t*, OCH_2CH_2 (npeoc)); 4.12–4.09 (*m*, MeO–C(6), H–C(4′)); 3.77 (*s*, 2 MeO); 3.38 (*m*, 2 H–C(5′)); 3.09 (*t*, OCH_2CH_2

(npeoc)); 2.79 (*m*, 1 H–C(2′)); 2.52 (*m*, 1 H–C(2′)). Anal. calc. for $C_{41}H_{40}N_6O_{10}\cdot H_2O$ (794.9): C 61.95, H 5.19, N 10.57; found: C 61.85, H 5.21, N 10.52.

23. *5′-O-(4,4′-Dimethoxytrityl)-O⁶-methylthymidine* (**43**). From **20** [22][23] by *G.P. A*. Purification by FC (AcOEt). Yield 84%. TLC ($CHCl_3$/MeOH 95:5): R_f 0.60. UV (MeOH): 281 (3.91), 278 (sh, 3.90), 230 (sh, 4.33). ^{1}H-NMR ($CDCl_3$): 7.86 (*s*, H–C(6)); 7.30 (*m*, 9 arom. H); 6.83 (*d*, 4 H *o* to MeO); 6.40 (*t*, H–C(1′)); 4.55 (*m*, H–C(3′)); 4.10 (*m*, H–C(4′)); 3.97 (*s*, MeO–C(4)); 3.79 (*s*, 2 MeO); 3.43 (*m*, 2 H–C(5′)); 2.64 (*d*, OH–C(3′)); 2.50 (*m*, 1 H–C(2′)); 2.27 (*m*, 1 H–C(2′)); 1.56 (*s*, Me–C(5)). Anal. calc. for $C_{32}H_{34}N_2O_7$ (558.6): C 68.80, H 6.13, N 5.01; found: C 68.14, H 6.45, N 4.75.

24. *5′-O-(4,4′-Dimethoxytrityl)-O⁶-ethylthymidine* (**44**) [26][27]. From **21** [22] by *G.P. A*. Purification by FC (AcOEt). Yield 91%. TLC ($CHCl_3$/MeOH 95:5): R_f 0.50. UV (MeOH): 281 (3.93), 278 (sh, 3.92), 230 (sh, 4.35). ^{1}H-NMR ($CDCl_3$): 7.82 (*s*, H–C(6)); 7.30 (*m*, 9 arom. H); 6.80 (*d*, 4 H *o* to MeO); 6.40 (*t*, H–C(1′)); 4.54 (*m*, H–C(3′)); 4.44 (*q*, MeCH_2O–C(4)); 4.09 (*m*, H–C(4′)); 3.80 (*s*, 2 MeO); 3.43 (*m*, 2 H–C(5′)); 2.61 (*m*, 1 H–C(2′)); 2.24 (*m*, 1 H–C(2′)); 2.19 (*d*, OH–C(3′)); 1.57 (*s*, Me–C(5)); 1.36 (*t*, *Me*CH_2O–C(4)). Anal. calc. for $C_{33}H_{35}N_2O_7$ (572.7): C 69.21, H 6.34, N 4.89; found: C 68.63, H 6.43, N 4.70.

25. *5′-O-(4,4′-Dimethoxytrityl)-O⁴-isopropylthymidine* (**45**). From **22** [22] by *G.P. A*. Purification by FC (AcOEt). Yield 85%. TLC ($CHCl_3$/MeOH 95:5): R_f 0.65. UV (MeOH): 281 (3.93), 278 (sh, 3.92), 230 (sh, 4.35). ^{1}H-NMR ($CDCl_3$): 7.82 (*s*, H–C(6)); 7.30 (*m*, 9 arom. H); 6.83 (*d*, 4 H *o* to MeO); 6.42 (*t*, H–C(1′)); 5.49 (m, Me_2C*H*O–C(4)); 4.55 (*m*, H–C(3′)); 4.10 (*m*, H–C(4′)); 3.79 (*s*, 2 MeO); 3.44 (*m*, 2 H–C(5′)); 2.63 (*m*, 1 H–C(2′), OH–C(3′)); 2.26 (*m*, 1 H–C(2′)); 1.54 (*s*, Me–C(5)); 1.32 (*m*, *Me*$_2$CHO–C(4)). Anal. calc. for $C_{34}H_{38}N_2O_7$ (586.7): C 69.61, H 6.53, N 4.77; found: C 69.09, H 6.73, N 4.58.

26. *O⁴-Butyl-5′-O-(4,4′-dimethoxytrityl)thymidine* (**46**). From **24** by *G.P. A*. Purification by FC (AcOEt). Yield 85%. TLC ($CHCl_3$/MeOH 95:5): R_f 0.65. UV (MeOH): 281 (3.94), 230 (sh, 4.34). ^{1}H-NMR ($CDCl_3$): 7.85 (*s*, H–C(6)); 7.42–7.22 (*m*, 9 arom. H); 6.83 (*d*, 4 H *o* to MeO); 6.42 (*t*, H–C(1′)); 4.56 (*m*, H–C(3′)); 4.36 (*t*, MeCH_2CH_2CH_2O); 4.12 (*m*, H–C(4′)); 3.79 (*s*, 2 MeO); 3.47 (*m*, 2 H–C(5′)); 3.00 (*m*, OH–C(3′)); 2.63 (*m*, 1 H–C(2′)); 2.26 (*m*, 1 H–C(2′)); 1.70 (*m*, MeCH_2CH_2CH_2O); 1.55 (*s*, Me–C(5)); 1.42 (*m*, MeCH_2CH_2CH_2O); 0.94 (*t*, *Me*CH_2CH_2CH_2O). Anal. calc. for $C_{35}H_{40}N_2O_7$ (600.7): C 69.98, H 6.71, N 4.66; found: C 69.63, H 6.43, N 4.70.

27. *5′-O-(4,4′-Dimethoxytrityl)-O⁴-[2-(4-nitrophenyl)ethyl]thymidine* (**47**). From **24** [22] by the *G.P. A*. Purification by FC ($CHCl_3$/MeOH 100:2). Yield 95%. TLC ($CHCl_3$/MeOH 95:5): R_f 0.82. UV (MeOH): 280 (sh, 4.25), 275 (4.26), 232 (sh, 4.40). ^{1}H-NMR ($CDCl_3$): 8.14 (*d*, 2 H *o* to NO_2); 7.92 (*s*, H–C(6)); 7.30 (*m*, 9 arom. H); 6.81 (*d*, 4 H *o* to MeO); 6.40 (*t*, H–C(1′)); 4.61 (*m*, H–C(3′), OCH_2CH_2 (npe)); 4.16 (*m*, H–C(4′)); 3.77 (*s*, 2 MeO); 3.73 (*d*, OH–C(3′)); 3.41 (*m*, 2 H–C(5′)); 3.15 (*t*, OCH_2CH_2 (npe)); 2.66 (*m*, 1 H–C(2′); 2.23 (*m*, 1 H–C(2′)); 1.46 (*s*, Me–C(5)). Anal. calc. for $C_{39}H_{38}N_3O_9\cdot 2H_2O$ (728.7): C 64.28, H 5.81, N 5.76; found: C 64.30, H 5.46, N 5.81.

28. *General Procedure B (G.P. B): Synthesis of 5′-O-(4,4′-Dimethoxytrityl)- and Base-Protected 3′-(2-Cyanoethyl Diisopropylphosphoramidites)* **52**–**66**. *Method I:* A mixture of protected nucleoside **35**–**40**, **43**–**45**, **47** (1 mmol), 2-cyanoethyl diisopropylphosphoramidochloridite (**48**) [28][29] (1.2 mmol) and ^{i}Pr$_2$EtN (0.69 ml, 3.79 mmol) in acid-free dry CH_2Cl_2 (4 ml) was stirred at r.t under Ar for 30 min (TLC control). The soln. was then diluted with CH_2Cl_2 (100 ml) and extracted with sat. $NaHCO_3$ soln. (2 × 50 ml), the aq. phase reextracted with CH_2Cl_2, the combined org. phase dried (Na_2SO_4) and evaporated, and the residue purified by CC or FC (silica gel) using the appropriate solvent systems to give the product as a diastereomer mixture which was co-evaporated with CH_2Cl_2: solid foam.

Method II: A mixture of protected nucleoside **33**, **34**, **41**, **42**, **46** (1 mmol), 2-cyanoethyl tetraisopropylphosphordiamidite (**49**) [30–33] (1.5 mmol), and 1*H*-tetrazole (0.05 mmol) in acid-free CH_2Cl_2 (13 ml) was stirred at r.t under Ar for 2 1/2 h. Workup as described above in *Method I*.

29. *2′-Deoxy-5′-O-(4,4′-dimethoxytrityl)-N⁶-[2-(4-nitrophenyl)ethoxycarbonyl]adenosine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**52**). From **33** by *G.P. B, Method II*. FC (toluene/AcOEt 3:7 and 1:1). Yield 92%. TLC (hexane/AcOEt/Et_3N 3:7:1): R_f 0.25, 0.17. UV (MeOH): 275 (sh, 4.41), 266 (4.47), 236 (4.45). ^{1}H-NMR ($CDCl_3$): 8.68 (*s*, H–C(8)); 8.25–8.15 (*m*, 2 H *o* to NO_2); 8.01 (*m*, NH); 7.36–7.34 (*d*, 2 H *m* to NO_2); 7.35–7.18 (*m*, 11 arom. H); 6.74 (*d*, 4 H *o* to MeO); 6.45 (*t*, H–C(1′)); 4.74 (*m*, H–C(3′)); 4.51 (*t*, OCH_2CH_2 (npeoc)); 4.29 (*m*, H–C(4′)); 3.75 (2 *s*, *m*, OCH_2CH_2CN, 2 MeO); 3.52–3.43 (*m*, 2 Me_2C*H*N); 3.31 (*m*, 2 H–C(5′)); 3.13 (*t*, OCH_2CH_2 (npeoc)); 2.82 (*m*, 1 H–C(2′)); 2.60 (*m*, 1 H–C(2′)); 2.59, 2.44 (2 *t*, OCH_2CH_2CN); 1.27–1.08 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 149.6, 149.5. Anal. calc. for $C_{49}H_{55}N_8O_{10}P$ (947.0): C 62.14, H 5.85, N 11.83; found: C 61.93, H 6.06, N 11.32.

30. *2′-Deoxy-5′-O-(4,4′-dimethoxytrityl)-N⁶-[(2-(4-nitrophenyl)ethoxycarbonyl]cytidine 3′-O-(2-Cyanoethyl Diisopropylphosphoramidite*) (**53**). From **34** [41] by *G.P. B, Method II*. FC (toluene/AcOEt 2:1 and 1:1).

Yield 85%. TLC (hexane/AcOEt/Et_3N 3:7:1): R_f 0.24, 0.13. UV (MeOH): 280 (sh, 4.40), 274 (4.42), 236 (4.56). ^{1}H-NMR ($CDCl_3$): 8.20 (*m*, 2 H *o* to NO_2); 8.20 (2 *s*, H–C(6)); 7.51–7.12 (*m*, 11 H, H *m* to NO_2, H *m* to MeO, Ar); 6.74 (*d*, 4 H *o* to OMe); 6.35 (*m*, H–C(1′), H–C(5)); 4.80 (*t*, OCH_2CH_2); 4.80 (*m*, H–C(3′)); 4.43 (*t*, OCH_2CH_2 (npeoc)); 4.21 (*m*, H–C(4′)); 3.90 (*m*, OCH_2CH_2 (npeoc)); 3.73 (2 *s*, 2 MeO); 3.70–3.47 (*m*, 2 Me_2CHN, OCH_2CH_2CN); 3.32–3.30 (2 *m*, 2 H–C(5′)); 3.08 (*m*, OCH_2CH_2 (npeoc)); 2.92 (*m*, 1 H–C(2′)); 2.75 (*m*, 1 H–C(2′)); 2.60, 224 (2 *t*, OCH_2CH_2CN); 1.10–0.91 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 150.0; 149.9. Anal. calc. for $C_{48}H_{55}N_6O_{11}P$ (923.0): C 62.46, H 6.00, N 9.10; found: C 62.46, H 6.08, N 8.83.

31. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*N^2*-2-[(4-nitrophenyl)ethyoxycarbonyl]guanosine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**54**). From **35** by *G.P. B*, *Method I*. CC (CH_2Cl_2/MeOH/Et_3N 97.5:2:0.5 → 91.5:8:0.5). Yield 60%. TLC ($CHCl_3$/MeOH/Et_3N 95:5:2): R_f 0.52. UV(MeOH): 281 (sh, 4.31), 272 (4.37), 258 (4.42), 249 (4.42), 237 (4.46). ^{1}H-NMR ($CDCl_3$): 8.15–8.11 (*m*, 2 H *o* to NO_2); 7.76, 7.72 (2 *s*, H–C(8)); 7.38–7.18 (*m*, 11 H, H *m* to NO_2, H *m* to MeO, Ar); 6.77–6.71 (*d*, 4 H *o* to MeO); 6.16 (*m*, H–C(1′)); 4.74–4.61 (*m*, H–C(3′)); 4.44 (*t*, OCH_2CH_2 (npeoc)); 4.24 (*m*, H–C(4′)); 3.74, 3.73 (2 *s*, 2 MeO); 4.14–3.45 (*m*, OCH_2CH_2CN, 2 Me_2CHN); 3.34–3.20 (*m*, 2 H–C(5′)); 3.09 (*t*, OCH_2CH_2 (npeoc)); 2.81–2.32 (*m*, 2 H–C(2′), OCH_2CH_2CN); 1.30–1.06 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 149.48; 148.89. Anal. calc. for $C_{49}H_{55}N_8O_{11}P$ (963.0): C 61.12, H 5.76, N 11.64; found: C 60.80, H 5.90, N 11.40.

32. *2′-Deoxy-5′-*O*-(4,4′dimethoxytrityl)-*N^2*-isobutyryl-*O^6*-2′-[2-(4-nitrophenyl)ethyl]guanosine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**55**). From **36** [18][19] by *G.P. B*, *Method I*. FC (AcOEt/Et_3N 99:1). Yield 70%. TLC (AcOEt/Et_3N 99:1): R_f 0.80, 0.72. UV (MeOH): 280 (sh, 4.53), 270 (4.47), 235 (4.43). ^{1}H-NMR ($CDCl_3$): 8.15–8.11 (*m*, H_o to NO_2); 7.98, 7.97 (2 *s*, H–C(8)); 7.81, 7.67 (2 *s*, H–N(2)); 7.49 (*d*, 2 H *m* to NO_2); 7.42–7.14 (*m*, 9 H, H *m* to MeO, Ar); 6.80–6,73 (*d*, 4 H *o* to MeO); 6.36 (*m*, H–C(1′)); 4.83–4.66 (*m*, *t*, H–C(3′), OCH_2CH_2 (npe)); 4.24 (*m*, H–C(4′)); 3.74 (*s*, 2 MeO); 3.88–3.49 (*m*, 2 Me_2CHN, OCH_2CH_2CN); 3.43–3.27 (*m*, 2 H–C(5′), OCH_2CH_2 (npe)); 2.89–2.50 (*m*, 2 H–C(2′), Me_2CHCO); 2.62–2.41 (2 *t*, OCH_2CH_2CN); 1.30–0.96 (*m*, 3 Me_2CH)). ^{31}P-NMR ($CDCl_3$): 149.6. Anal. calc. for $C_{52}H_{61}N_8O_{10}P$ (989.1): C 63.15, H 6.22, N 11.33; found: C 62.58, H 6.35, N 11.04.

33. *2′-Deoxy-5′-*O*-(4,4′dimethoxytrityl)-*N^2*-[2-(4-nitrophenyl)ethoxycarbonyl]-*O^6*-[2-(4-nitrophenyl)ethyl]-guanosine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**56**). From **37** by *G.P. B*, *Method I*. CC (AcOEt/Et_3N 99:1). Yield 86%. TLC (AcOEt/Et_3N 99:1): R_f 0.81, 0.76. UV (MeOH): 269 (4.55), 236 (4.48). ^{1}H-NMR ($CDCl_3$): 8.17–8.11 (*m*, 2 H *o* to NO_2); 7.96, 7.95 (2 *s*, H–C(8)); 7.51–7.13 (*m*, 11 arom. H); 6.74 (*d*, 4 H *o* to MeO); 6.36 (*m*, H–C(1′)); 4.82–4.70 (*m*, *t*, 3 H–C(3′), OCH_2CH_2 (npe)); 4.42 (*t*, OCH_2CH_2 (npeoc)); 4.24 (*m*, H–C(4′)); 3.74 (2 *s*, 2 MeO); 3.86–3.48 (*m*, 2 Me_2CHN, OCH_2CH_2CN); 3.37–3.26 (*m*, 2 H–C(5′), OCH_2CH_2 (npe)); 3.09 (*t*, OCH_2CH_2 (npeoc)); 2.87–2.55 (*m*, 2 H–C(2′)); 2.62, 2.41 (2 *t*, OCH_2CH_2CN); 1.26–1.07 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 149.59; 149.38. Anal. calc. for $C_{57}H_{62}N_9O_{13}P$ (1112.2): C 61.56, H 5.62, N 11.33; found: C 61.76, H 5.65, N 11.05.

34. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*N^2*-isobutyryl-*O^6*-methylguanosine 3′-(2-Cyanoethyl Diisopropyl-phosphoramidite)* (**57**). From **38** [27] by *G.P. B*, *Method I*. FC (AcOEt/Et_3N 96:4). Yield 91%. TLC (AcOEt/Et_3N 96:4): R_f 0.71. UV(MeOH): 280 (sh, 4.09), 269 (4.25), 234 (sh, 4.36), 221 (sh, 4.52). ^{1}H-NMR ($CDCl_3$): 8.01 (*s*, H–C(8)); 7.73 (*br. s*, NH); 7.45–7.20 (*m*, 9 arom. H); 6.80 (*d*, 4 H *o* to MeO); 6.42 (*t*, H–C(1′)); 4.73 (*m*, H–C(3′)); 4.18 (*s*, MeO–C(4)); 3.80 (*s*, 2 MeO); 3.89–3.57 (*m*, 2 Me_2CHN, OCH_2CH_2CN); 3.38 (*m*, 2 H–C(5′)); 3.20 (*m*, Me_2CHCO); 2.90 (*m*, 1 H–C(2′)); 2.64 (*m*, *t*, 2 H, OCH_2CH_2CN, 1 H–C(2′)); 2.50 (*t*, 1 H, OCH_2CH_2CN); 1.33–1.13 (*m*, 3 Me_2CH)). ^{31}P-NMR ($CDCl_3$): 149.44; 149.37. Anal. calc. for $C_{45}H_{56}N_7O_8P \cdot H_2O$ (871.9): C 61.98, H 6.70, N 11.24; found: C 61.81, H 6.65, N 11.22.

35. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*O^6*-ethyl-*N^2*-isobutyrylguanosine 3′-(2-Cyanoethyl Diisopropyl-phosphoramidite)* (**58**). From **39** [27] by *G.P. B*, *Method I*. FC (AcOEt/Et_3N 96:4). Yield 91%. TLC (AcOEt/Et_3N 96:4): R_f 0.75. UV(MeOH): 280 (sh, 4.10), 269 (4.26), 234 (sh, 4.37), 221 (sh, 4.54). ^{1}H-NMR ($CDCl_3$): 7.95, 7.93 (2 *s*, H–C(8)); 7.74, 7.67 (2 *s*, NH); 7.41–7.5 (*m*, 9 arom. H); 6.77–6.73 (*d*, 4 H *o* to MeO); 6.40 (*m*, H–C(1′)); 4.75 (*m*, H–C(3′)); 4.60 (*q*, $MeCH_2O$–C(6)); 4.27 (*m*, H–C(4′)); 3.75 (2 *s*, 2 MeO); 3.83–3.44 (*m*, 2 Me_2CHN, OCH_2CH_2CN); 3.35–3.30 (*m*, 2 H–C(5′)); 3.10 (*m*, Me_2CHCO); 2.82 (*m*, 1 H–C(2′)); 2.64 (*m*, *t*, 2 H, OCH_2CH_2CN, 1 H–C(2′)); 2.42 (*t*, 1 H, OCH_2CH_2CN); 1.48 (*t*, $MeCH_2O$–C(6)); 1.26–1.04 (*m*, 3 Me_2CH). ^{31}P-NMR ($CDCl_3$): 149.46; 149.39. Anal. calc. for $C_{46}H_{58}N_7O_8P \cdot 1.5\ H_2O$ (895.0): C 61.73, H 6.87, N 10.95; found: C 61.70, H 6.98, N 10.93.

36. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*O^6*-isopropyl-*N^2*-isobutyrylguanosine 3′-(2-Cyanoethyl Diisopro-pylphosphoramidite)* (**59**). From **40** [27] by *G.P. B*, *Method I*. FC (toluene/AcOEt 2:1 → 1:1). Yield 93%. TLC (toluene/AcOEt 2:1): R_f 0.7. UV (MeOH): 280 (sh, 4.10), 269 (4.26), 233 (sh, 4.36), 221 (sh, 4.53). ^{1}H-NMR ($CDCl_3$): 8.02 (2 *s*, H–C(8)); 7.86, 7.79 (2 *s*, NH); 7.46–7.21 (*m*, 9 arom. H); 6.83–6.79 (*d*, 4 H *o* to MeO); 6.42 (*t*, H–C(1′)); 5.64–5.59 (*m*, Me_2CHO–C(6)); 4.83 (*m*, H–C(3′)); 4.32 (*m*, H–C(4′)); 3.79–3.60 (*m*, *s*,

2 Me_2CHN, 2 MeO, 1 H of CH_2CH_2CN); 3.30–3.26 (*m*, 2 H–C(5′)); 3.10 (*m*, Me_2CHCO); 2.90 (*m*, 1 H–C(2′)); 2.67 (*t*, *m*, 1 H of CH_2CH_2CN, 1 H–C(2′)); 2.42 (*t*, 1 H, OCH_2CH_2CN); 1.50 (*m*, Me_2CHCO); 1.27–1.15 (*m*, Me_2CHO–C(6)); 1.15–1.02 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 149.47; 149.36. Anal. calc. for $C_{47}H_{60}N_7O_8P \cdot 1.5H_2O$ (909.0): C 62.10, H 6.98, N 10.79; found: C 61.86, H 7.00, N 10.83.

37. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*O^6*-butyl-*N^2*-isobutyrylguanosine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**60**). From **41** [27] by *G.P. B*, *Method I*. FC (toluene/AcOEt 2:1 → 1:1). Yield 72%. TLC (toluene/AcOEt 2:1): R_f 0.7. UV (MeOH): 280 (sh, 4.12), 269 (4.27), 233 (sh, 4.37). ^{1}H-NMR ($CDCl_3$): 7.95, 7.93 (2 *s*, H–C(8)); 7.71, 7.64 (2 *s*, H–N(2)); 7.35–7.13 (*m*, 9 arom. H); 6.77–6.73 (*d*, 4 H *o* to MeO); 6.31 (*t*, H–C(1′)); 4.68 (*m*, H–C(3′)); 4.47 (*t*, $MeCH_2CH_2CH_2O$); 4.19 (*m*, H–C(4′)); 3.69 (2 *s*, 2 MeO); 3.58 (*m*, 2 Me_2CHN, OCH_2CH_2CN); 3.30–3.26 (*m*, 2 H–C(5′)); 3.10 (*m*, Me_2CHCO); 2.72 (*m*, 1 H–C(2′)); 2.57 (*t*, *m*, 2 H, OCH_2CH_2CN, 1 H–C(2′)); 2.37 (*t*, 1 H, OCH_2CH_2CN); 1.78 (*m*, $MeCH_2CH_2CH_2O$); 1.46 (*m*, $MeCH_2CH_2CH_2O$); 1.17–1.02 (*m*, 3 Me_2CH); 0.90 (*t*, $MeCH_2CH_2CH_2O$). ^{31}P-NMR ($CDCl_3$): 149.59; 149.46. Anal. calc. for $C_{48}H_{62}N_7O_8P$ (895.9): C 64.34, H 6.98, N 10.94; found: C 63.63, H 7.00, N 10.65.

38. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*O^6*-methyl-*N^2*-[2-(4-nitrophenyl)ethoxycarbonyl]guanosine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**61**). From **42** by *G.P. B*, *Method I*. FC (toluene/AcOEt 1:1). Yield 76%. TLC (AcOEt): R_f 0.43. UV (MeOH): 268 (4.13), 236 (4.15). ^{1}H-NMR ($CDCl_3$): 8.18 (*d*, 2 H *o* to NO_2); 7.91, 7.89 (2 *s*, H–C(8)); 7.53–7.19 (*m*, 11 arom. H); 6.77–6.74 (*d*, 4 H *o* to MeO); 6.38 (*t*, H–C(1′)); 4.74 (*m*, H–C(3′)); 4.44 (*t*, OCH_2CH_2 (npeoc)); 4.26 (*m*, H–C(4′)); 4.14 (*s*, MeO–C(6)); 3.76 (2 *s*, 2 MeO); 3.68 (*m*, OCH_2CH_2CN, 2 Me_2CHN); 3.38 (*m*, 2 H–C(5′)); 3.11 (*t*, OCH_2CH_2 (npeoc)); 2.87 (*m*, 1 H–C(2′)); 2.61 (*t*, *m*, 2 H, 1 H–C(2′)), OCH_2CH_2CN); 2.44 (*t*, 1 H, OCH_2CH_2CN); 1.22–1.08 (*m*, 2 Me_2CH). ^{31}P-NMR ($CDCl_3$): 149.42; 149.28. Anal. calc. for $C_{50}H_{57}N_8O_{11}P$ (977.0): C 61.46, H 5.88, N 11.47; found: C 61.10, H 5.87, N 10.87.

39. *5′-*O*-(4,4′-Dimethoxytrityl)-*O^4*-methylthymidine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**62**). From **43** [22] [23] by *G.P. B*, *Method I*. CC (AcOEt/Et_3N 199:1). Yield 73%. TLC (AcOEt/Et_3N 199:1): R_f 0.77, 0.71. UV (MeOH): 281 (3.92), 276 (3.93), 232 (4.33). ^{1}H-NMR ($CDCl_3$): 7.95, 7.92 (2 *s*, H–C(6)); 7.42–7.19 (2 *m*, 9 arom. H); 6.80 (*dd*, 4 H *o* to MeO); 6.42 (*m*, H–C(1′)); 4.64 (*m*, H–C(3′)); 4.16 (*m*, H–C(4′)); 3.97 (*s*, MeO–C(4)); 3.77, 3.76 (2 *s*, 7 H, 2 MeO, OCH_2CH_2CN); 3.60–3.26 (*m*, 4 H, 1 H–C(5′), 2 Me_2CHN, OCH_2CH_2CN); 3.30 (*m*, 1 H–C(5′)); 2.65–2.56 (*m*, *t*, 2 H, OCH_2CH_2CN, 1 H–C(2′)); 2.39 (*t*, 1 H, OCH_2CH_2CN); 1.49 (2 *s*, Me–C(5)); 1.17–1.02 (*m*, *d*, 2 Me_2CH). ^{31}P-NMR ($CDCl_3$): 149.81; 149.21. Anal. calc. for $C_{41}H_{51}N_4O_8P$ (758.8): C 64.89, H 6.77, N 7.38; found: C 63.85, H 6.81, N 7.53.

40. *5′-*O*-(4,4′-Dimethoxytrityl)-*O^4*-ethylthymidine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**63**). From **44** by *G.P. B, Method I*. CC (AcOEt/Et_3N 199:1). Yield 80%. TLC (AcOEt/Et_3N 199:1): R_f 0.76, 0.72. UV (MeOH) 281 (3.95), 276 (3.94), 232 (4.35). ^{1}H-NMR ($CDCl_3$): 7.90, 7.84 (2 *s*, H–C(6)); 7.42–7.19 (2 *m*, 9 arom. H); 6.82–6.79 (*dd*, 4 H *o* to MeO); 6.42 (*m*, H–C(1′)); 4.64 (*m*, H–C(3′)); 4.16 (*m*, H–C(4′)); 4.44 (*q*, $MeCH_2MeO$–C(4)); 3.77, 3.76 (2 *s*, 7 H, 2 MeO, OCH_2CH_2CN); 3.60–3.26 (*m*, 4 H, 1 H–C(5′), 2 Me_2CH, OCH_2CH_2CN); 3.30 (*m*, 1 H–C5(′)); 2.65–2.56 (*m*, *t*, 2 H, OCH_2CH_2CN, 1 H–C(2′)); 2.34 (*t*, 1 H, OCH_2CH_2CN); 2.25 (*m*, 1 H–C(2′)); 1.49, 1.47 (2 *s*, Me–C(5)); 1.35 (*t*, $MeCH_2O$–C(6)); 1.17–0.99 (*m*, 2 Me_2CH). ^{31}P-NMR ($CDCl_3$): 149.80; 149.21. Anal. calc. for $C_{42}H_{53}N_4O_8P$ (772.8): C 65.27, H 6.91, N 7.25; found: C 64.42, H 6.83, N 7.31.

41. *5′-*O*-(4,4′-Dimethoxytrityl)-*O^4*-isopropylthymidine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**64**). From **45** by *G.P. B*, *Method I*. CC (AcOEt/Et_3N 199:1). Yield 77%. TLC (AcOEt/Et_3N 199:1): R_f 0.87, 0.85. UV (MeOH) 281 (3.96), 276 (3.95), 232 (4.34). ^{1}H-NMR ($CDCl_3$): 7.85, 7.78 (2 *s*, H–C(6)); 7.42–7.19 (2 *m*, 9 arom. H); 6.80 (*dd*, 4 H *o* to MeO); 6.42 (*m*, H–C(1′)); 5.47 (*m*, Me_2CHO–C(4)); 4.64 (*m*, H–C(3′)); 4.16 (*m*, H–C(4′)); 3.77, 3.76 (2 *s*, 7 H, 2 MeO, OCH_2CH_2CN)); 3.60–3.26 (*m*, 4 H, OCH_2CH_2CN, 1 H–C(5′), 2 Me_2CHN); 3.28 (*m*, 1 H–C(5′)); 2.65–2.56 (*m*, *t*, 2 H, OCH_2CH_2CN, 1 H–C(2′)); 2.34 (*t*, 1 H, OCH_2CH_2CN); 2.25 (*m*, 1 H–C(2′)); 1.45, 1.43 (2 *s*, Me–C(5)); 1.29 (*m*, Me_2CHO–C(4)); 1.17–0.99 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 149.84; 149.28. Anal. calc. for $C_{43}H_{56}N_4O_8P$ (786.87): C 65.63, H 7.05, N 7.12; found: C 64.33, H 7.11, N 7.24.

42. *O^4-Butyl-5′-*O*-(4,4′-dimethoxytrityl)thymidine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**65**). From **46** [27] by *G.P. B*, *Method II*. CC (toluene/AcOEt 1:1). Yield 82%. TLC (toluene/AcOEt 1:1): R_f 0.58, 0.50. UV (MeOH): 281 (3.95), 276 (3.95), 226 (4.39). ^{1}H-NMR ($CDCl_3$): 7.85, 7.78 (2 *s*, H–C(6)); 7.42–7.19 (2 *m*, 9 arom. H); 6.80 (*dd*, 4 H *o* to MeO); 6.42 (*m*, H–C(1′)); 4.64 (*m*, H–C(3′)); 4.36 (*t*, $MeCH_2CH_2CH_2O$); 4.16 (*m*, H–C(4′)); 3.79 (2 *s*, 7 H, 2 MeO, OCH_2CH_2CN); 3.60–3.40 (*m*, 4 H, 1 H–C(5′), 2 Me_2CHN, OCH_2CH_2CN); 3.30 (*m*, 1 H–C(5′)); 2.65 (*m*, *t*, 2 H, 1 H–C(2′), OCH_2CH_2CN); 2.39 (*t*, 1 H, OCH_2CH_2CN); 2.27 (*m*, 1 H–C(2′)); 1.71 (*m*, $MeCH_2CH_2CH_2O$); 1.46 (*m*, *d*, Me–C(5), $MeCH_2CH_2CH_2O$); 1.17–0.99 (*d*, *m*, 2 Me_2CHN); 0.94 (*t*, $MeCH_2CH_2CH_2O$). ^{31}P-NMR ($CDCl_3$): 148.50; 147.23. Anal. calc. for $C_{44}H_{57}N_4O_8P$ (800.8): C 65.98, H 7.17, N 7.00; found: C 65.70, H 7.22, N 7.08.

43. *5′-O-(4,4′-Dimethoxytrityl)-O⁴-[2-(4-nitrophenyl)ethyl]thymidine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**66**). From **47** by *G.P. B*, *Method I*. FC (toluene/AcOEt 1:1). Yield 74%. TLC (toluene/AcOEt 1:1): R_f 0.42. UV (MeOH): 280 (sh, 4.24), 275 (4.25), 232 (sh, 4.39). ^{1}H-NMR ($CDCl_3$): 8.16 (*d*, 2 H *o* to NO_2); 7.90 (*d*, H–C(6)); 7.30 (2 *m*, 11 arom. H); 6.82 (*dd*, 4 H *o* to MeO); 6.38 (*m*, H–C(1′)); 4.63 (*m*, H–C(3′), OCH_2CH_2 (npe)); 4.15 (*m*, H–C(4′)); 3.78 (2 *s*, 7 H, 2 MeO, OCH_2CH_2CN); 3.54 (*m*, 4 H, 2 Me_2CHN, OCH_2CH_2CN, 1 H–C(5′)); 3.32 (*m*, 1 H–C(5′)); 3.16 (*t*, OCH_2CH_2 (npe)); 2.64 (*m*, *t*, 2 H, 1 H–C(2′), OCH_2CH_2CN); 2.32 (*m*, *t*, 2 H, 1 H–C(2′), OCH_2CH_2CN); 1.41 (*d*, Me–C(5)); 1.11 (*d*, *m*, 2 Me_2CH). ^{31}P-NMR ($CDCl_3$): 149.71; 149.16. Anal. calc. for $C_{48}H_{56}N_5O_{12}P$ (894.0): C 64.49, H 6.31, N 7.83; found: C 64.45, H 6.54, N 7.31.

44. *General Procedure C: Synthesis of 5′-O-(4,4′-Dimethoxytrityl)- and Base-Protected 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidites]* **67**–**82**. *Method I:* A mixture of the protected nucleoside (1 mmol), 2-(4-nitrophenyl)ethyl diisopropylphosphoramidochloridite (**50**) [39] (1.2 mmol) and ^{i}Pr$_2$EtN (0.69 ml, 3.79 mmol) in acid-free dry CH_2Cl_2 (4 ml) was stirred at r.t under Ar for 30 min (TLC control). The soln. was diluted with CH_2Cl_2 (100 ml) and extracted with sat. $NaHCO_3$ soln. (2 × 50 ml), the aq. phase reextracted with CH_2Cl_2, the combined org. layer dried (Na_2SO_4) and evaporated, and the residue purified by CC or FC (silica gel) using the appropriate solvent systems to give the product as a diastereomer mixture which was co-evaporated with CH_2Cl_2: solid foam.

Method II: Analogously to *Method I*, with 2-(4-nitrophenyl)ethyl tetraisopropylphosphorodiamidite (**51**) [39] (1.5 mmol) and 1*H*-tetrazole (0.05 mmol) in acid-free CH_2Cl_2 (13 ml) by stirring at r.t. under Ar for 2 1/2 h.

45. *5′-O-(4-Monomethoxytrityl)thymidine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**67**). From **25** [15] by *G.P. C*, *Method I*. CC (AcOEt/Et_3N 95:5). Yield 62%. TLC (AcOEt/Et_3N 95:5): R_f 0.72. UV (MeOH): 269 (4.30), 234 (sh, 4.25). ^{1}H-NMR ($CDCl_3$): 8.63 (*br. s*, NH); 8.21 (*m*, 2 H *o* to NO_2); 7.61 (*m*, H–C(6)); 7.42–7.21 (*m*, 14 arom. H); 6.75 (*m*, 2 H *o* to MeO); 6.43 (*m*, H–C(1′)); 4.64 (*m*, H–C(3′)); 4.16 (*m*, H–C(4′)); 3.79 (2 *s*, MeO); 3.93–3.62 (*m*, OCH_2CH_2 (npe)); 3.60–3.26 (*m*, 2 H–C(5′), 2 Me_2CHN); 3.05–2.82 (2 *t*, OCH_2CH_2 (npe)); 2.51–2.17 (*m*, 2 H–C(2′); 1.42 (*s*, Me–C(5)); 1.17–0.99 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 147.49. Anal. calc. for $C_{44}H_{51}N_4O_9P$ (810.9): C 65.17, H 6.34, N 6.91; found: C 64.06, H 6.50, N 6.89.

46. *2′-Deoxy-5′-O-(4-monomethoxytrityl)-N⁶-[2-(4-nitrophenyl)ethoxycarbonyl]adenosine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**68**). From **26** [12] by *G.P. C*, *Method I*. FC (AcOEt/Et_3N 96:4). Yield 66%. TLC (AcOEt/Et_3N 95:5): R_f 0.65. UV (MeOH): 275 (sh, 4.52), 268 (4.56), 236 (4.34). ^{1}H-NMR ($CDCl_3$): 8.69–8.68 (*2 s*, H–C(8)); 8.31 (*br. s*, NH); 8.21–8.07 (*m*, H–C(2), 4 H *o* to NO_2); 7.45–7.16 (*m*, 16 arom. H); 6.77 (*m*, 2 H *o* to MeO); 6.45 (*m*, H–C(1′)); 4.70 (*m*, H–C(3′)); 4.53 (*t*, OCH_2CH_2 (npeoc)); 4.35 (*m*, H–C(4′)); 3.93–3.77 (*m*, OCH_2CH_2 (PO-npe)); 3.76 (2 *s*, MeO); 3.69–3.47 (*m*, 2 Me_2CHN); 3.45–3.27 (*m*, 2 H–C(5′)); 3.15 (*t*, OCH_2CH_2 (npeoc)); 3.04–2.87 (*2 t*, OCH_2CH_2 (PO-npe)); 2.87 (*m*, 1 H–C(2′)); 2.57 (*m*, 1 H–C(2′)); 1.21–1.06 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 147.67. Anal. calc. for $C_{53}H_{57}N_8O_{11}P$ (1013.0): C 62.84, H 5.67, N 11.06; found: C 62.33, H 5.82, N 10.76.

47. *2′-Deoxy-5′-O-(4-monomethoxytrityl)-N⁴-[2-(4-nitrophenyl)ethoxycarbonyl]cytidine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**69**). From **27** [12] by *G.P. C*, *Method I*. FC (AcOEt/Et_3N 96:4). Yield 72%. TLC (AcOEt/Et_3N 95:5): R_f 0.69. UV (MeOH): 282 (4.40), 276 (4.41), 236 (4.47). ^{1}H-NMR ($CDCl_3$): 8.26–8.20 (*m*, 4 H *o* to NO_2, H–C(6)); 7.60 (*br. s*, NH); 7.44–7.21 (*m*, 16 arom. H); 6.93–6.81 (*m*, 2 H *o* to MeO, H–C(5)); 6.25 (*m*, H–C(1′)); 4.55 (*m*, H–C(3′)); 4.43 (*t*, OCH_2CH_2 (npeoc)); 4.17 (*m*, H–C(4′)); 3.95–3.64 (*m*, OCH_2CH_2 (PO-npe)); 3.73 (2 *s*, MeO); 3.60–3.32 (*m*, 2 Me_2CHN, 2 H–C(5′)); 3.14–3.08 (*t*, OCH_2CH_2 (npeoc)); 3.03–2.85 (2 *t*, OCH_2CH_2 (PO-npe)); 2.72 (*m*, 1 H–C(2′)); 2.29 (*m*, 1 H–C(2′)); 1.27–1.01 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 147.85. Anal. calc. for $C_{52}H_{57}N_6O_{12}P$ (989.0): C 63.15, H 5.81, N 8.50; found: C 61.19, H 5.93, N 8.31.

48. *2′-Deoxy-5′-O-(monomethoxytrityl)-N²-[2-(4-nitrophenyl)ethoxycarbonyl]-O⁶-[2-(4-nitrophenyl)ethyl]-guanosine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**70**). From **28** [12] by *G.P. C*, *Method I*. CC (AcOEt/Et_3N 96:4). Yield 78%. TLC (AcOEt/Et_3N 96:4): R_f 0.75, 0.67. UV (MeOH): 270 (4.62), 236 (4.42). ^{1}H-NMR ($CDCl_3$): 8.22–8.07 (*m*, 6 H *o* to NO_2); 7.96, 7.95 (2 *s*, H–C(8)); 7.54–7.18 (*m*, 18 arom. H); 6.88–6.74 (*m*, 2 H *o* to MeO); 6.39 (*m*, H–C(1′)); 4.82 (*t*, OCH_2CH_2 (npe)); 4.65 (*m*, H–C(3′)); 4.45 (*t*, OCH_2CH_2 (npeoc)); 4.21 (*m*, H–C(4′)); 3.90 (*m*, OCH_2CH_2 (PO-npe)); 3.76 (2 *s*, MeO); 3.61–3.43 (*m*, 2 Me_2CHN); 3.38–3.30 (2 *m*, 2 H–C(5′), OCH_2CH_2 (PO-npe)); 3.10 (*t*, OCH_2CH_2 (npeoc)); 3.00–2.82 (2 *t*, OCH_2CH_2 (PO-npe)); 2.84–2.51 (2 *m*, 2 H–C(2′)); 1.27–1.03 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 147.85. Anal. calc. for $C_{61}H_{64}N_9O_{14}P$ (1178.2): C 62.19, H 5.48, N 10.70; found: C 61.62, H 5.64, N 10.64.

49. *5′-O-(4,4′-Dimethoxytrityl)thymidine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**71**). From **29** [15] by *G.P. C*, *Method I*. FC (toluene/AcOEt/Et_3N 70:30:1 → 50:50:1). Yield 82%. By *G.P. C*, *Method II*. FC (toluene/AcOEt 2:1 → 1:1). Yield 76%. TLC (AcOEt): R_f 0.87. UV (MeOH): 269 (4.32), 234

(4.38). ^{1}H-NMR ($CDCl_3$): 8.29 (*s*, NH); 8.13–8.05 (*m*, 2 H *o* to NO_2); 7.59 (*m*, H–C(6)); 7.38–7.21 (*m*, 11 arom. H); 6.80 (*m*, 4 H *o* to MeO); 6.38 (*m*, H–C(1′)); 4.55 (*m*, H–C(3′)); 4.12 (*m*, H–C(4′)); 3.75 (2 *s*, *m*, 2 MeO, OCH_2CH_2 (PO-npe)); 3.61 (*m*, 1 H–C(5′), 2 Me_2CHN); 3.32 (*m*, 1 H–C(5′)); 2.97, 2.82 (2 *t*, OCH_2CH_2 (PO-npe)); 2.31–2.21 (2 *m*, 2 H–C(2′)); 1.38 (*s*, Me–C(5)); 1.23–0.99 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 148.49; 148.09. Anal. calc. for $C_{45}H_{54}N_4O_{10}P$ (841.9): C 64.18, H 6.46, N 6.65; found: C 63.31, H 6.25, N 6.59.

50. *N^6-Benzoyl-2′-deoxy-5′-*O*-(4,4′-dimethoxytrityl)adenosine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**72**). From **30** [15] by *G.P. C*, *Method II*. FC (petroleum ether/acetone 3:1 and 2:1). Yield 65%. TLC (AcOEt): R_f 0.70. UV (MeOH): 272 (4.48), 232 (4.49). ^{1}H-NMR ($CDCl_3$): 9.01 (*s*, NH); 8.03 (2 *s*, H–C(8)); 8.16–8.00 (*m*, H–C(2), 2 H *o* to NO_2); 7.78–7.50 (*m*, 2 H *o* to CO); 7.40–7.21 (*m*, 14 arom. H); 6.95–6.75 (*d*, 4 H *o* to MeO); 6.48 (*d*, H–C(1′)); 4.91–4.83 (*m*, H–C(3′)); 4.50 (*m*, H–C(4′)); 3.77 (*s*, 2 MeO); 3.91–3.62 (*m*, OCH_2CH_2 (PO-npe)); 3.54–3.33 (2 *m*, 2 H–C(5′), 2 Me_2CHN); 2.90, 2.60 (2 *t*, *m*, OCH_2CH_2 (PO-npe), 1 H–C(2′)); 2.46 (*m*, 1 H–C(2′)); 1.42–0.84 (*m*, 2 Me_2CHN). Anal. calc. for $C_{52}H_{56}N_7O_9P$ (954.0): C 65.46, H 5.91, N 10.27; found: C 64.93, H 5.88, N 9.70.

51. *N^4-Benzoyl-2′-deoxy-5′-*O*-(4,4′-dimethoxytrityl)cytidine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**73**). From **31** [15] by *G.P. C*, *Method II*. FC (toluene/AcOEt 2:1 and 1:1). TLC (AcOEt): R_f 0.72. UV (MeOH): 305 (4.04), 260 (4.50), 234 (4.53). ^{1}H-NMR ($CDCl_3$): 8.46 (*s*, NH); 8.15–8.05 (*dd*, H–C(6)); 7.93 (*m*, 2 H *o* to CO); 7.80–7.70 (*m*, 2 H *o* to NO_2); 7.68–7.53 (*m*, 14 arom. H); 6.99–6.82 (*d*, 4 H *o* to MeO); 6.27 (*d*, H–C(1′)); 4.42 (*m*, H–C(3′)); 4.19 (*m*, H–C(4′)); 3.80 (2 *s*, *m*, 2 MeO, OCH_2CH_2); 3.70 (2 *m*, 2 H–C(5′), 2 Me_2CHN); 3.18, 2.87 (2 *t*, OCH_2CH_2); 2.76 (*m*, 1 H–C(2′)); 2.32 (*m*, 1 H–C(2′)); 1.15–1.02 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 148.81; 148.47. Anal. calc. for $C_{51}H_{56}N_5O_{10}P$ (930.0): C 65.86, H 6.06, N 7.53; found: C 65.73, H 6.01, N 6.91.

52. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-N^2-isobutyrylguanosine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**74**). From **32** [42] by *G.P. C*, *Method I*. FC ($AcOEt/Et_3N$ 100:1). Yield 85%. TLC ($AcOEt/Et_3N$ 99:1): R_f 0.45. UV (MeOH): 281 (sh, 4.36), 273 (4.39), 261 (4.42), 253 (sh, 4.41), 236 (4.44). ^{1}H-NMR ($CDCl_3$): 11.93 (*s*, NH); 8.11 (*m*, 2 H *o* to NO_2); 7.81–7.70 (*m*, H–C(8), NH); 7.48–7.17 (*m*, 11 arom. H); 6.78–6.73 (*m*, 4 H *o* to MeO); 6.13 (*m*, H–C(1′)); 4.64 (*m*, H–C(3′)); 4.16 (*m*, H–C(4′)); 3.88 (*m*, OCH_2CH_2 (PO-npe)); 3.74–3.72 (3 *s*, 2 MeO); 3.50–3.10 (2 *m*, 2 H–C(5′), 2 Me_2CHN); 3.00–2.82 (*m*, Me_2CHCO, OCH_2CH_2 (PO-npe)); 2.42 (*m*, 1 H–C(2′)); 1.84 (*m*, 1 H–C(2′)); 1.11–0.84 (*m*, 3 Me_2CH). Anal. calc. for $C_{49}H_{58}N_7O_{10}P$ (936.0): C 62.88, H 6.25, N 10.47; found: C 62.46, H 6.37, N 10.10.

53. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-N^6-[2-(4-nitrophenyl)ethoxycarbonyl]adenosine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**75**). From **33** by *G.P. C.*, *Method I*. FC (toluene/AcOEt/NEt_3 70:30:1 → 50:50:1). Yield 87%. TLC (AcOEt): R_f 0.66. UV (MeOH): 274 (sh, 4.55), 267 (4.58), 236 (4.45). ^{1}H-NMR ($CDCl_3$): 8.68 (*s*, H–C(8)); 8.31 (*br. s*, NH); 8.25 (*s*, H–C(2)); 8.15 (*m*, 4 H *o* to NO_2); 7.42–7.12 (*m*, 13 arom. H); 6.75 (*d*, 4 H *o* to MeO); 6.35 (*t*, H–C(1′)); 4.65 (*m*, H–C(3′)); 4.42 (*t*, OCH_2CH_2 (npeoc)); 4.21 (*m*, H–C(4′)); 3.90–3.70 (*m*, OCH_2CH_2 (PO-npe)); 3.73 (2 *s*, 2 MeO); 3.52–3.43 (*m*, 2 Me_2CHN); 3.31 (2 *m*, 2 H–C(5′)); 3.08 (*t*, OCH_2CH_2 (npeoc)); 3.00–2.82 (2 *t*, *m*, OCH_2CH_2 (PO-npe), 1 H–C(2′)); 2.62 (*m*, 1 H–C(2′)); 1.25–1.02 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 148.67; 148.37. Anal. calc. for $C_{54}H_{59}N_8O_{12}P$ (1043.1): C 62.18, H 5.70, N 10.74; found: C 61.72, H 6.07, N 9.96.

54. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-N^4-[2-(4-nitrophenyl)ethoxycarbonyl]cytidine 3′-[2-(4-Nitrophenyl)ethyl Düsopropylphosphoramidite]* (**76**). From **34** [41] *G.P. C*, *Method I*. FC ($AcOEt/Et_3N$ 96:4). Yield 91%. By *G.P. C*, *Method II*. FC (toluene/AcOEt 2:1 and 1:1). Yield 66%. TLC ($AcOEt/Et_3N$ 96:4): R_f 0.68. UV (MeOH): 282 (sh, 4.41), 276 (4.41), 236 (4.56). ^{1}H-NMR ($CDCl_3$): 8.46 (*s*, NH); 8.19 (*d*, H–C(6)); 8.14–8.04 (*dd*, 4 H *o* to NO_2); 7.42–7.02 (*m*, 13 arom. H); 6.84 (*d*, H–C(5), 4 H *o* to MeO); 6.23 (*m*, H–C(1′)); 4.58 (*m*, H–C(3′)); 4.42 (*t*, OCH_2CH_2 (npeoc)); 4.15 (*m*, H–C(4′)); 3.88–3.61 (2 *s*, *m*, 2 MeO, OCH_2CH_2 (PO-npe)); 3.47 (2 *m*, 2 H–C(5′), 2 Me_2CHN); 3.10 (*t*, OCH_2CH_2 (npeoc)); 2.87 (2 *t*, OCH_2CH_2 (PO-npe)); 2.69 (*m*, 1 H–C(2′)); 2.18 (*m*, 1 H–C(2′)); 1.20–1.01 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 148.81; 148.47. Anal. calc. for $C_{53}H_{60}N_6O_{13}P$ (1019.0): C 62.46, H 5.83, N 8.24; found: C 61.24, H 5.81, N 8.00.

55. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-N^2-isobutyryl-O^6-[2-(4-nitrophenyl)ethyl]guanosine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**77**). From **36** [18] by *G.P. C*, *Method I*. FC (toluene/AcOEt/Et_3N 70:30:1 → 50:50:1). Yield 85%. TLC ($AcOEt/Et_3N$ 99:1): R_f 0.79. UV (MeOH): 280 (sh, 4.50), 270 (4.58), 235 (4.47). ^{1}H-NMR ($CDCl_3$): 8.18–8.05 (*m*, 4 H *o* to NO_2); 7.97, 7.95 (2 *s*, H–C(8)); 7.66, 7.64 (2 *s*, NH); 7.51–7.12 (*m*, 13 arom. H); 6.74 (*d*, 4 H *o* to MeO); 6.34 (*m*, H–C(1′)); 4.83–4.78 (*t*, OCH_2CH_2 (CO-npe)); 4.68 (*m*, H–C(3′)); 4.21 (*m*, H–C(4′)); 3.88 (*m*, OCH_2CH_2 (PO-npe)); 3.73 (2 *s*, 2 MeO); 3.59–3.41 (*m*, 2 Me_2CHN); 3.36–3.22 (2 *m*, 2 H–C(5′), OCH_2CH_2 (CO-npe)); 3.01 (*t*, 1 H, OCH_2CH_2 (PO-npe)); 2.92–2.68 (*t*, *m*, 3 H, 1 H–C(2′), Me_2CHCO, OCH_2CH_2 (PO-npe)); 2.48 (*m*, 1 H–C(2′)); 1.29–1.02 (*m*, 3 Me_2CH). ^{31}P-

NMR ($CDCl_3$): 148.21; 148.56. Anal. calc. for $C_{57}H_{65}N_8O_{12}P$ (1085.2): C 63.09, H 6.04, N 10.33; found: C 61.36, H 6.09, N 10.02.

56. *2′-Deoxy-5′-*O*-(4,4′-dimethoxytrityl)-*N^2*-[2-(4-nitrophenyl)ethoxycarbonyl]-*O^6*-[2-(4-nitrophenyl)-ethyl]guanosine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**78**). From **37** by *G.P. C*, *Method I*. FC (AcOEt/Et_3N 96:4). Yield 91%. By *G.P. C*, *Method II*. FC (toluene/AcOEt 2:1 and 1:1). Yield 68%. TLC (AcOEt/Et_3N 96:4): R_f 0.83. UV (MeOH): 269 (4.60), 237 (4.46). ^{1}H-NMR ($CDCl_3$): 8.17–8.05 (*m*, 6 H *o* to NO_2); 7.95 (2 *s*, H–C(8)); 7.51–7.14 (*m*, 15 arom. H); 6.75–6.72 (*d*, 4 H *o* to MeO); 6.35 (*m*, H–C(1′)); 4.80 (*t*, OCH_2CH_2 (CO-npe); 4.65 (*m*, H–C(3′)); 4.43 (*t*, OCH_2CH_2 (npeoc)); 4.20 (*m*, H–C(4′)); 3.90 (*m*, OCH_2CH (PO-npe)); 3.73 (2 *s*, 2 MeO); 3.52–3.43 (*m*, 2 Me_2CHN); 3.31 (2 *m*, 2 H–C(5′), OCH_2CH_2 (CO-npe)); 3.09 (*t*, OCH_2CH_2 (npeoc)); 3.00–2.84 (2 *t*, OCH_2CH_2 (PO-npe)); 2.82 (*m*, 1 H–C(2′)); 2.45 (*m*, 1 H–C(2′)); 1.25–1.02 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 148.37; 148.28. Anal. calc. for $C_{62}H_{66}N_9O_{15}P$ (1208.2): C 61.63, H 5.51, N 10.43; found: C 60.77, H 5.75, N 10.20.

57. *5′-*O*-(4,4′-Dimethoxytrityl)-*O^4*-methylthymidine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**79**). From **43** [27][28] by *G.P. C*, *Method I*. FC (toluene/AcOEt 1:2). Yield 73%. TLC (toluene/AcOEt 1:1): R_f 0.56. UV (MeOH): 281 (sh, 4.23), 275 (4.24), 234 (sh, 4.36). ^{1}H-NMR ($CDCl_3$): 8.10 (*m*, 2 H *o* to NO_2); 7.86 (*dd*, H–C(6)); 7.30 (*m*, 11 arom. H); 6.80 (*dd*, 4 H *o* to MeO); 6.38 (*m*, H–C(1′)); 4.57 (*m*, H–C(3′)); 4.13 (*m*, H–C(4′)); 3.98 (*s*, MeO); 3.75 (2 *s*, 2 MeO, OCH_2CH_2 (PO-npe)); 3.49 (*m*, 1 H–C(5′), 2 Me_2CHN); 3.28 (*m*, 1 H–C(5′)); 2.99, 2.84 (2 *t*, OCH_2CH_2 (PO-npe)); 2.60 (*m*, 1 H–C(2′)); 2.21 (*m*, 1 H–C(2′)); 1.50 (*s*, Me–C(5)); 1.06 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 148.31; 148.01. Anal. calc. for $C_{46}H_{55}N_4O_{10}P$ (854.9): C 64.63, H 6.48, N 6.55; found: C 63.94, H 6.35, N 6.31.

58. *5′-*O*-(4,4′-Dimethoxytrityl)-*O^4*-ethylthymidine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**80**). From **44** [22][26–29] by *G.P. C*, *Method I*. FC (toluene/AcOEt 1:2). Yield 71%. TLC (toluene/AcOEt 1:1): R_f 0.62. UV (MeOH): 280 (sh, 4.25), 275 (4.26), 232 (sh, 4.39). ^{1}H-NMR ($CDCl_3$): 8.10 (*m*, 2 H *o* to NO_2); 7.85 (*dd*, H–C(6)); 7.31 (2 *m*, 11 arom. H); 6.80 (*dd*, 4 H *o* to MeO); 6.39 (*m*, H–C(1′)); 4.51 (*m*, $MeCH_2O$–C(4), H–C(3′)); 4.13 (*m*, H–C(4′)); 3.75 (*m*, 2 MeO, OCH_2CH_2 (PO-npe)); 3.48 (*m*, 1 H–C(5′), 2 Me_2CHN); 3.28 (*m*, 1 H–C(5′)); 2.99, 2.84 (2 *t*, OCH_2CH_2 (PO-npe)); 2.62 (*m*, 1 H–C(2′)); 2.21 (*m*, 1 H–C(2′)); 1.49 (*s*, Me–C(5)); 1.36 (*m*, $MeCH_2O$–C(4)); 1.06 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 148.29; 148.00. Anal. calc. for $C_{47}H_{57}N_4O_{10}P$ (869.0): C 64.96, H 6.61, N 6.45; found: C 64.44, H 6.58, N 6.39.

59. *5′-*O*-(4,4′-Dimethoxytrityl)-*O^4*-isopropylthymidine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**81**). From **45** [27] by *G.P. C*, *Method I*. FC (toluene/AcOEt 1:1). Yield 71%. TLC (toluene/AcOEt 1:1): R_f 0.71. UV (MeOH): 280 (sh, 4.26), 275 (4.27), 232 (sh, 4.39). ^{1}H-NMR ($CDCl_3$): 8.10 (*m*, 2 H *o* to NO_2); 7.82 (*d*, H–C(6)); 7.29 (2 *m*, 11 arom. H); 6.81 (*d*, 4 H *o* to MeO); 6.41 (*m*, H–C(1′)); 5.50 (*m*, Me_2CHO–C(4)); 4.57 (*m*, H–C(3′)); 4.13 (*m*, H–C(4′)); 3.75 (*m*, 2 MeO, OCH_2CH_2 (PO-npe)); 3.49 (*m*, 1 H–C(5′), 2 Me_2CHN); 3.28 (*m*, 1 H–C(5′)); 2.99, 2.84 (2 *t*, OCH_2CH_2 (PO-npe)); 2.60 (*m*, 1 H–C(2′)); 2.22 (*m*, 1 H–C(2′)); 1.48 (*s*, Me–C(5)); 1.32 (*m*, Me_2CHO–C(4)); 1.06 (*m*, 2 Me_2CHN). ^{31}P-NMR ($CDCl_3$): 148.28; 147.98. Anal. calc. for $C_{48}H_{59}N_4O_{10}P$ (883.0): C 65.29, H 6.74, N 6.35; found: C 64.89, H 6.86, N 6.26.

60. *5′-*O*-(4,4′-Dimethoxytrityl)-*O^4*-2-[2-(4-nitrophenyl)ethyl]thymidine 3′-[2-(4-Nitrophenyl)ethyl Diisopropylphosphoramidite]* (**82**). From **47** by *G.P. C*, *Method II*. FC (AcOEt/Et_3N 99:1). Yield 88%. By *G.P. C*, *Method II*. FC (toluene/AcOEt 2:1 → 1:1). Yield 79%. TLC (AcOEt): R_f 0.77. UV (MeOH): 281 (sh, 4.30), 275 (4.32), 234 (sh, 4.33). ^{1}H-NMR ($CDCl_3$): 8.63 (*m*, 4 H *o* to NO_2); 7.61 (*m*, H–C(6)); 7.42–7.21 (*m*, 13 arom. H); 6.76 (*m*, 4 H *o* to MeO); 6.43 (*m*, H–C(1′)); 4.64 (*m*, H–C(3′), OCH_2CH_2 (npe)); 4.16 (*m*, H–C(4′)); 3.79 (2 *s*, 2 MeO); 3.93–3.62 (*m*, OCH_2CH_2 (PO-npe)); 3.60–3.26 (*m*, 2 H–C(5′), 2 Me_2CHN), OCH_2CH_2 (npe)); 3.05–2.82 (2 *t*, OCH_2CH_2 (PO-npe)); 2.51 (*m*, 1 H–C(2′)); 2.17 (*m*, 1 H–C(2′)); 1.42 (*s*, Me–C(5)); 1.17–0.99 (*m*, *2* Me_2CHN). ^{31}P-NMR ($CDCl_3$): 148.50; 147.23. Anal. calc. for $C_{53}H_{60}N_5O_{12}P$ (990.1): C 64.29, H 6.18, N 7.07; found: C 63.92, H 5.96, N 6.39.

REFERENCES

[1] M. Beier, W. Pfleiderer, *Helv. Chim. Acta* **1999**, *82*, 879.
[2] T. W. Greene, P. G. M. Wuts, 'Protective Groups in Organic Synthesis', 3rd edn., John Wiley & Sons, New York, 1999.
[3] V. Amarnath, A. D. Broom, *Chem. Rev.* **1977**, *77*, 183.
[4] J. Engels, E. Uhlmann, *Adv. Biochem. Eng. Biotechnol.* **1988**, *37*, 73.
[5] S. L. Beaucage, R. P. Iyer, *Tetrahedron* **1992**, *48*, 2223.
[6] S. L. Beaucage, R. P. Iyer, *Tetrahedron* **1993**, *49*, 1925.

[7] S. L. Beaucage, R. P. Iyer, *Tetrahedron* **1993**, *49*, 6123.
[8] R. L. Letsinger, V. Mahadevan, *J. Am. Chem. Soc.* **1965**, *87*, 3526.
[9] T. Brown, D. J. S. Brown, in 'Oligonucleotides and Analogues – A Practical Approach', Ed. F. Eckstein, IRL Press, Oxford, 1991, p. 1.
[10] M. A. Dorman, S. A. Noble, L. J. McBride, M. H. Caruthers, *Tetrahedron* **1984**, *40*, 95.
[11] H. Köster, J. Biernat, J. McManus, A. Wolter, A. Stumpe, C. K. Narang, N. D. Sinha, *Tetrahedron* **1984**, *40*, 103.
[12] F. Himmelsbach, B. S. Schulz, T. Trichtinger, R. Charubala, W. Pfleiderer, *Tetrahedron* **1984**, *40*, 59.
[13] K. P. Stengele, W. Pfleiderer, *Tetrahedron Lett.* **1990**, *31*, 2549.
[14] G. S. Ti, B. L. Gaffney, R. Jones, *J. Am. Chem. Soc.* **1982**, *104*, 1316.
[15] H. Schaller, G. Weimann, B. Lerch, H. G. Khorana, *J. Am. Chem. Soc.* **1963**, *85*, 3821.
[16] O. Mitsunobu, *Synthesis* **1981**, 1.
[17] B. S. Schulz, W. Pfleiderer, *Nucleosides Nucleotides* **1987**, *6*, 529.
[18] B. L. Gaffney, R. A. Jones, *Tetrahedron Lett.* **1982**, *23*, 2253, 2257.
[19] B. L. Gaffney, L. A. Marky, R. A. Jones, *Tetrahedron* **1984**, *40*, 3.
[20] H. Büchi, H. G. Khorana, *J. Mol. Biol.* **1972**, *72*, 251.
[21] B. S. Schulz, W. Pfleiderer, *Tetrahedron Lett.* **1983**, *24*, 3587.
[22] L. Kiriasis, S. Farkas, W. Pfleiderer, *Nucleosides Nucleotides* **1986**, *5*, 517.
[23] C. B. Reese, P. A. Skone, *J. Chem. Soc., Perkin Trans. 1* **1984**, 1263.
[24] W. L. Sung, *J. Org. Chem.* **1982**, *47*, 3623.
[25] B. L. Li, C. B. Reese, P. F. Swann, *Biochemistry* **1987**, *26*, 1086.
[26] P. K. Bridson, W. T. Markiewicz, C. B. Reese, *J. Chem. Soc., Chem. Commun.* **1977**, 791.
[27] H. Borowy-Borowsky, R. W. Chambers, *Biochemistry* **1987**, *26*, 2465.
[28] H. Borowy-Borowsky, R. W. Chambers, *Biochemistry* **1989**, *28*, 1471.
[29] D. F. Forner, Y. Palom, S. Ikuta, E. Pedroso, R. Eritja, *Nucleic Acids Res.* **1990**, *18*, 5729.
[30] N. D. Sinha, J. Biernat, H. Köster, *Nucleic Acids Res.* **1984**, *12*, 4539.
[31] G. Zon, K. A. Gallo, C. J. Samson, K. Shao, R. A. Byrd, M. F. Summers, *Nucleic Acids Res.* **1985**, *13*, 8181.
[32] A. D. Barone, J. Y. Tang, M. H. Caruthers, *Nucleic Acids Res.* **1984**, *12*, 4051.
[33] A. Kraszewski, K. E. Norris, *Nucleic Acids Res.* **1987**, *15*, 177.
[34] J. Nielsen, J. E. Marugg, M. Taagard, J. H. van Boom, O. Dahl, *Nucleic Acids Res.* **1986**, *14*, 4051.
[35] J. Nielsen, O. Dahl, *Nucleic Acids Res.* **1987**, *15*, 3626.
[36] E. Uhlmann, W. Pfleiderer, *Tetrahedron Lett*. **1980**, *21*, 481.
[37] W. Pfleiderer, M. Schwarz, H. Schirmeister, *Chem. Scripta* **1986**, *26*, 147.
[38] P. Herdewijn, R. Charubala, W. Pfleiderer, *Helv. Chim. Acta* **1989**, *72*, 1729.
[39] R. W. Sobol, E. E. Henderson, N. Kon, J. Shao, P. Hitzges, E. Mordechai, N. L. Reichenbach, R. Charubala, W. Pfleiderer, R. J. Suhadolnik, *J. Biol. Chem.* **1995**, *270*, 5963.
[40] E. Uhlmann, W. Pfleiderer, *Helv. Chim. Acta* **1981**, *64*, 1688.
[41] S. Waldvogel, W. Pfleiderer, *Helv. Chim. Acta* **1998**, *81*, 46.
[42] K. L. Agarwal, F. Riftina, *Nucleic Acids Res.* **1978**, *5*, 2805.

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