Oligonucleotides Containing Disaccharide Nucleosides

by **Ekaterina V. Efimtseva**[a]), **Georgii V. Bobkov**[a]), **Sergey N. Mikhailov**[a])[b]), **Arthur Van Aerschot**[b]), **Guy Schepers**[b]), **Roger Busson**[b]), **Jef Rozenski**[b]), **Piet Herdewijn**[b])

[a]) Engelhardt Institute of Molecular Biology, Russian Academy of Sciences, Vavilov street 32, Moscow, 119991 Russia (Tel. +7(95)1359704; fax +7(95)1351405; e-mail: smikh@genome.eimb.rssi.ru)

[b]) Rega Institute, Katholieke Universiteit Leuven, Minderbroedersstraat 10, B-3000 Leuven (Tel. +32-16-337387; fax +32-16-337340; e-mail: Piet.Herdewijn@rega.kuleuven.ac.be)

Disaccharide nucleosides with 2′-*O*-(D-arabinofuranosyl), 2′-*O*-(L-arabinofuranosyl), 2′-*O*-(D-ribopyranosyl), 2′-*O*-(D-erythrofuranosyl), and 2′-*O*-(5-azido-5-deoxy-D-ribofuranosyl) substituents were synthesized. These modified nucleosides were incorporated into oligonucleotides (see *Table*). Single substitution resulted in a ΔT_m of $+0.5$ to $-1.4°$ for DNA/RNA and a ΔT_m of -0.8 to $-4.7°$ for DNA/DNA duplexes. These disaccharide nucleosides can be well accommodated in RNA/DNA duplexes, and the presence of a $NH_2-C(5'')$ group has a beneficial effect on duplex stability.

1. Introduction. – Modification at the 2′-*O*-position of the carbohydrate moiety of ribonucleic acids, has proven to be a successful approach for oligonucleotide modification in the antisense technology [1–3]. The introduction of a 2′-*O*-(methoxyethyl) (MOE) substituent increases nuclease resistance, and MOE-oligonucleotides surpass the high RNA-binding affinity of 2′-*O*-methylribonucleosides [1–3].

A large number of oligonucleotides containing an aminoalkyl modification at the sugar moiety have been reported, the amino group normally being protonated at physiological pH. Oligonucleotides having such a positively charged modification exhibit high binding properties towards RNA and DNA and maintain nuclease resistance [4–6]. Among the 2′-*O*-substituents developed, recently high affinity was found for the aminoethyl [4], aminopropyl [5], and (dimethylamino)propyl [6] derivatives which may form specific contact with a proximal phosphate group in the nucleic-acids duplex. Because of its natural occurrence [7], we studied the effect of the introduction of an additional sugar moiety at the 2′-position of natural ribonucleosides into RNA. A furanose sugar might indeed fill the minor groove and replace part of the hydration shell. Thus, 2′-*O*-(β-D-ribofuranosyl)nucleosides were synthesized and incorporated into synthetic oligonucleotides [8–12]. The solution structure of the RNA duplex containing 2′-*O*-(β-D-ribofuranosyl)adenosine was determined by NMR spectroscopy [12], demonstrating that the RNA duplex maintains an A-type helix with all ribose residues adopting a C(3′)-*endo* conformation. It was thus shown that this kind of 2′-*O*-modified adenosine has no profound effect on the RNA structure and on the thermal stability of the duplex, the additional ribofuranosyl moiety taking up a well-defined position in the minor groove [12].

These results encouraged us to synthesize novel disaccharide nucleosides containing different furanose and pyranose residues at the 2′-*O*-position and incorporate them into oligonucleotides to improve the affinity towards complementary RNA and DNA.

Likewise, the synthesis of a disaccharide nucleoside with a 5-amino-5-deoxyribofuranosyl moiety was developed with the aim of introducing a positive charge at the sugar moiety. The influence of the different 2′-*O*-substituents on the stability of duplex formation was evaluated.

2. Results and Discussion. – 2.1. *Synthesis.* The synthesis of the disaccharide nucleosides **2a**–**e** (see *Fig.*) was performed according to previously described procedures. It consists in the condensation of *N*,3′-*O*,5′-*O*-protected ribonucleosides **1** and 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-*β*-D-ribofuranose in the presence of $SnCl_4$ in dichloroethane [8], conditions originally proposed for the preparation of alkyl *β*-D-ribofuranosides [13]. Sugars that were successfully used in this reaction are peracylated D- (and L-) arabinofuranoses, D-ribopyranose, and D-erythrofuranose.

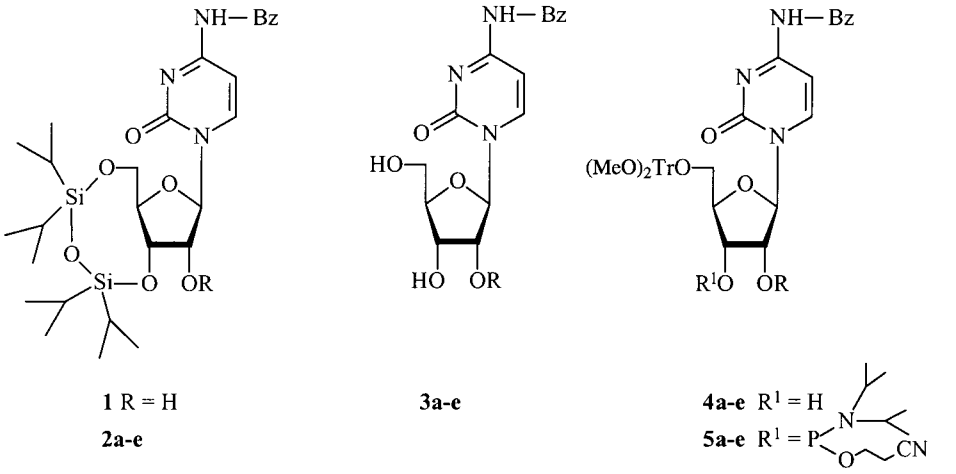


Figure. *Structure of disaccharide nucleosides and their phosphoramidite derivatives used for oligonucleotide synthesis*

The synthesis of disaccharide nucleoside **2a** was described previously [8]. Analogous condensation of **1** with an excess of 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-D- or L-arabinofuranose [14] in the presence of $SnCl_4$ for 16 h at 0° gave disaccharides **2b** and **2c** in good yields. Pyranoses are usually less reactive [15][16] than furanoses under similar conditions. The reaction between nucleoside **1** and 1,2,3,4-tetra-*O*-acetyl-*β*-D-ribopyranose proceeded more slowly (3 days at 0°); nevertheless the yield of **2d** was 86%. The disaccharide nucleoside **2e** was prepared from 1,2,3-tri-*O*-acetyl-D-erythrose

[17] in 51% yield. The selective deblocking of the silyl group gave partially protected **3b**–**e**, which were converted by standard procedures to the corresponding dimethoxytrityl derivatives **4b**–**e** and their phosphoramidites **5b**–**e**. The preparation of phosphoramidite **5a** was reported previously [10][11].

The structure of the compounds was confirmed by NMR spectroscopy. The *O*-glycosylation reaction proceeded stereospecifically with the formation of 1″,2″-*trans* substituted disaccharide nucleosides **2b**–**e**. The coupling constants ($J(1'',2'')$) in the additional furanose moiety for compounds **2a**–**c** and **2e** are less than 0.5 Hz. Only for the ribopyranosyl derivative **2d** is $J(1'',2'')$ 2.3 Hz. The assignments of the protons and C-atoms in the ^{1}H- and ^{13}C-NMR spectra were made in analogy with related disaccharide nucleosides [8][16].

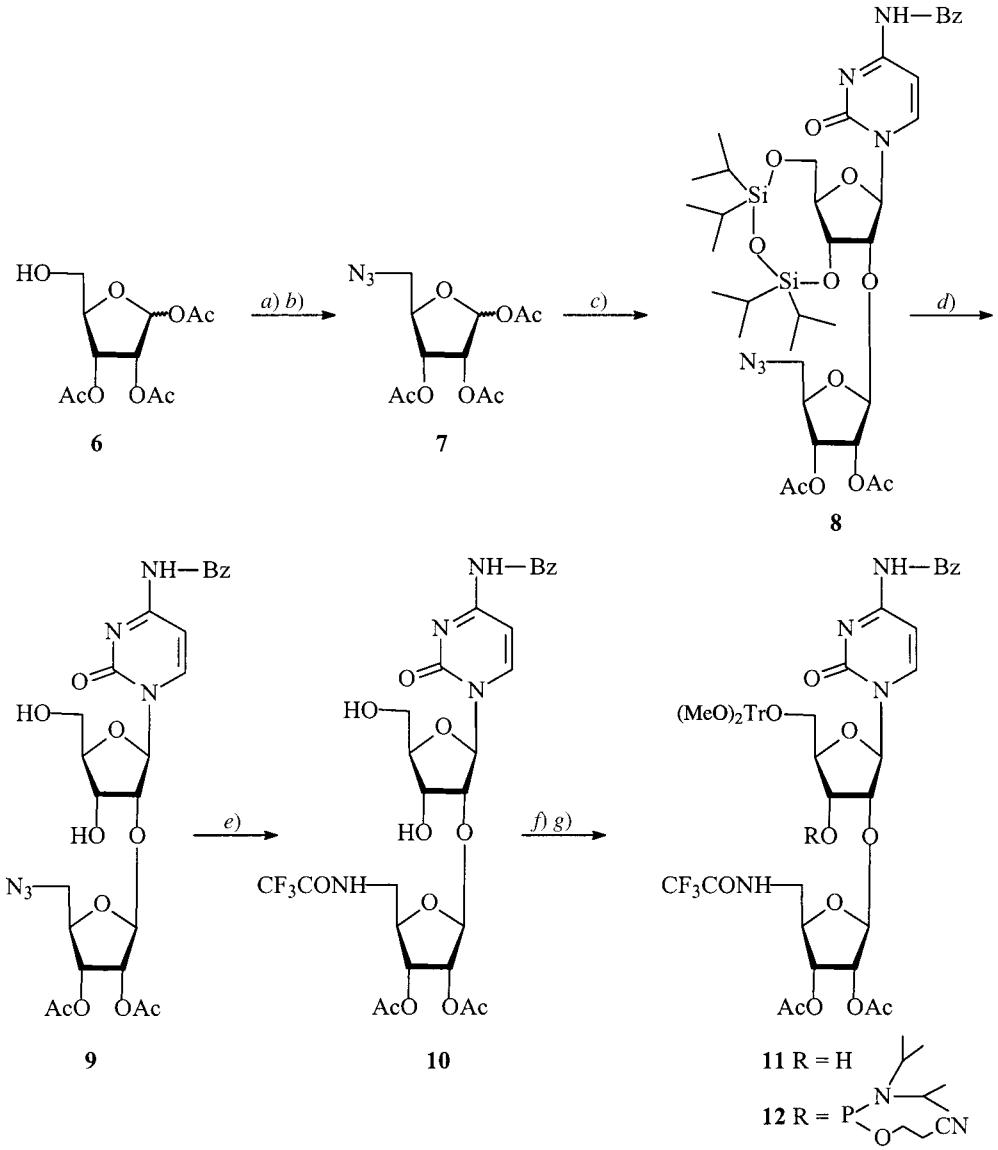
The synthesis of amino-sugars is often performed *via* azido derivatives, which are reduced in a strategic step during synthesis. The readily available triacetate **6** was chosen as the starting material [18] (*Scheme*). Introduction of the azide functionality consisted in sulfonylation of the primary OH group with tosyl chloride followed by replacement with azide ion [19][20], without chromatographic separation of the intermediate 5-*O*-tosylated sugar. Treatment of the crude intermediate with NaN_3 in DMF afforded azide **7** in 77% yield (*Scheme*). The condensation of ribonucleoside **1** with **7** under standard conditions gave disaccharide nucleoside **8**. After desilylation, nucleoside **9** was obtained in good yield.

Different methods have been used for the conversion of an azide to an amino group: for example, hydrogenolysis, reduction with metal hydrides, sulfide ions, or tin derivatives, reaction with phosphines, and others [21][22]. Treatment with PPh_3 followed by alkaline hydrolysis [23] was inconvenient due to the presence of acyl-protecting groups in the disaccharide nucleoside **9**. The reaction with H_2S in pyridine [24][25] was incomplete and resulted in low yield. Reduction with Bu_3SnH in the presence of 2,2′-azobis[isobutyronitrile] (AIBN) in dioxane was more successful [26]. The main difficulty encountered with this conversion concerned the chromatographic purification of the free amino derivative due to its polar character and the possible migration of an *O*-acyl group. Such intramolecular migration under reducing conditions was observed previously [26]. To avoid the formation of an undesirable *N*-acyl by-product, simultaneous *N*-(trifluoroacetylation) was carried out. One-step reduction of **9** in the presence of an excess of ethyl trifluoroacetate afforded **10** in 59% yield after silica gel chromatography. The obtained analogue **10** was converted *via* **11** to the phosphoramidite **12** in good overall yield. The structure of the compounds containing azido and amino groups were established by NMR and mass spectroscopy.

The ^{13}C-NMR spectra show a high-field shift for the C(5′) signal (at 53 ppm) of the ribosyl residue of disaccharides **8** and **9**, indicative of the presence of azide. The ^{1}H-NMR spectra of nucleoside **10** clearly indicates the presence of the CF_3CONH group, with the NH moiety appearing as a *dd* at 8.79 ppm with $J(NH,5'') = 5.8$ and 6.3 Hz. The signals at 158.2 ppm ($J(C{=}O,F) = 37$ Hz) and 116.0 ppm ($J(C,F) = 288$ Hz) in the ^{13}C-NMR spectrum confirm the presence of the CF_3CO group in **10**. A high-field shift for the C(5′) signal of the ribosyl residue (41.76 ppm) is observed.

2.2. *Oligonucleotide Synthesis and Characterization.* The phosphoramidites **5a**–**e** and **12** were incorporated into several oligonucleotides on an automated DNA synthesizer. Two series of dodecamers with sequence 5′-d(GCATAT**X**A**X**TGG)-3′

Scheme



a) TsCl, Py, $ClCH_2CH_2Cl$. *b*) NaN_3, DMF, 80°. *c*) **1**, $SnCl_4$, $ClCH_2CH_2Cl$. *d*) Bu_3NF; THF. *e*) Bu_3SnH, AIBN, CF_3COOEt, dioxane, 90°. *f*) $(MeO)_2TrCl$, Py. *g*) $(^iPr_2N)_2P(OCH_2CH_2CN)$, 1*H*-tetrazole.

were obtained with either a single or double substitution by a disaccharide nucleoside analogue at the **X** position, and the correct composition for all analogue-containing sequences was confirmed by ESI-MS (*Table*). The binding affinities of the modified oligonucleotides towards complementary RNA and DNA were evaluated. Thus, melting temperatures (T_m) were determined in 0.1M NaCl-containing buffer, and these

are listed in the *Table*. The T_m values of the unmodified reference DNA/RNA and DNA/DNA duplexes were 49 and 49.5°, respectively. All modified oligonucleotides containing disaccharide nucleoside analogues displayed higher binding affinity towards complementary RNA *vs.* DNA sequences. However, the thermal stability decreased more pronouncedly upon introduction of a second analogue. The ΔT_m, compared to the reference duplexes, ranged from -0.7 to $-2.2°$ for the DNA/RNA and from -3.7 to $-10.4°$ for the DNA/DNA interactions, respectively. A single substitution exerted only marginal influence on the T_m values, with ΔT_m ranging from -1.4 to $+0.5°$ for DNA/RNA and from -0.8 to $-4.7°$ for the DNA/DNA duplexes. The incorporation of the larger, more constrained 2′-*O*-(β-D-ribopyranosyl)cytidine analogue into oligonucleotides resulted in the largest destabilization of RNA and DNA duplexes. In contrast, a single substitution with the amine-containing nucleoside analogue obtained from **12** slightly increased the binding affinity of the thus modified oligonucleotide for the RNA complementary sequence.

Table. *Molecular-Mass Measurements and Melting Experiments of Oligodeoxyribonucleotides Containing Disaccharide Nucleosides*

X[a])	5′-d(GCATAT**X**ACTGG)-3′				5′-d(GCATAT**X**A**X**TGG)-3′			
	calc. mi mass[b])	found mass	T_m[c]) RNA compl.	T_m[c]) DNA compl.	calc. mi mass[b])	found mass	T_m[c]) RNA compl.	T_m[c] DNA compl.
dCyd	3643.65	3643.90	49.0	49.5	3643.65	3643.90	49.0	49.5
β-D-RibFur-Cyd	3791.69	3791.98	49.3	44.9	3939.73	3940.04	48.4	40.1
β-D-RibPyr-Cyd	3791.69	3791.98	48.6	45.2	3939.73	3940.06	46.8	39.1
α-D-AraFur-Cyd	3791.69	3792.24	49.0	46.5	3939.73	3940.32	46.9	41.5
α-L-AraFur-Cyd	3791.69	3792.26	49.0	46.2	3939.73	3940.32	46.8	40.8
β-D-EryFur-Cyd	3761.68	3761.98	48.9	44.8	3879.70	3880.00	47.5	39.7
β-D-5′NH_2RibFur-Cyd	3790.70	3790.92	49.5	48.7	3937.76	3938.02	48.3	45.8

[a]) **X**: dCyd = 2′-deoxycytidine, β-D-RibFur-Cyd = 2′-*O*-(β-D-ribofuranosyl)cytidine, β-D-RibPyr-Cyd = 2′-*O*-(β-D-ribopyranosyl)cytidine, α-D-AraFur-Cyd = 2′-*O*-(α-D-arabinofuranosyl)cytidine, α-L-AraFur-Cyd = 2′-*O*-(α-L-arabinofuranosyl)cytidine, β-D-EryFur-Cyd = 2′-*O*-(β-D-erythrofuranosyl)cytidine, and β-D-5′NH_2RibFur-Cyd = 2′-*O*-(5-amino-5-deoxy-β-D-ribofuranosyl)cytidine unit. [b]) Calculated monoisotopic mass. [c]) Melting temperatures (°, at 260 nm) determined in 0.1M NaCl, 20 mM KH_2PO_4, pH 7.5, 0.1 mM EDTA at a concentration of 4 μM of each oligonucleotide.

3. Conclusion. – Several new versatile disaccharide synthons, useful for oligonucleotide synthesis, were synthesized under the form of 2′-*O*-glycosylated nucleoside analogues. Incorporation of such sterically demanding substituents at the 2′-*O*-position do not have a profound effect on the thermal stability of the corresponding duplexes. The decrease in T_m is more pronounced in DNA/DNA duplexes that in RNA/DNA duplexes. The presence of an $NH_2-C(5'')$ function has a beneficial effect on duplex stability. The additional diol groups of the sugar moiety can be easily oxidized with sodium periodate to yield oligonucleotide derivatives with dialdehyde moieties. It was shown before that such oligonucleotides might be used for affinity labelling of different enzymes involved in nucleic-acid biosynthesis [10][11].

Experimental Part

General. Column chromatography (CC): silica gel (0.06–0.20 mm); FC = flash chromatography. TLC: silica gel *260 F* (*Merck*); eluents: CH_2Cl_2 (*A*); CH_2Cl_2/MeOH 98:2 (*B*); CH_2Cl_2/MeOH 95:5 (*C*); CH_2Cl_2/MeOH 9:1 (*D*); hexane/Et_2O 1:2 (*E*); detection by UV light. NMR Spectra: *Bruker AMX-400* and *Varian Unity 500* NMR spectrometer; at 300 K; chemical shifts δ in ppm rel. to the solvent signals (^1H and ^{13}C) and rel. to the external ref. H_3PO_4 (capil.) (^{31}P); coupling constants *J* in Hz. The signals were assigned by double-resonance techniques and COSY experiments. Mass spectrometry and exact mass measurements: quadrupole/orthogonal-acceleration time-of-flight tandem mass spectrometer (*Q-Tof-2*, *Micromass*, Manchester, UK), equipped with a standard electrospray-ionization (ESI) interface; the composition of the modified oligonucleotides was verified by ESI-MS analysis of samples infused in ^{i}PrOH/H_2O 1:1 at 3 µl/min LSI-MS. (Liquid secondary-ion mass spectra): *Kratos Concept-1 H* mass spectrometer (Manchester, UK).

Oligonucleotide Synthesis. Oligonucleotide synthesis was performed on an *ABI 392A-DNA* synthesizer (*Applied Biosystems*) by means of the phosphoramidite approach. The standard 1-µmol scale DNA assembly protocol was used, except for a 15 min coupling time with 0.12M of the incoming amidite for the modified analogues. The oligomers were deprotected and cleaved from the solid support by treatment with conc. aq. ammonia (50°, 16 h). Following gel filtration on a *NAP-10*® column (*Sephadex G25*, DNA grade, *Pharmacia*), purification was achieved on a *Mono-Q*®*-HR-10/10* anion-exchange column (*Pharmacia*) with the following gradient system: *E* = 10 mM NaOH, pH 12.0, 0.1M NaCl; *G* = 10 mM NaOH, pH 12.0, 0.9M NaCl; the applied gradient depended on the oligomer; flow rate 2 ml min^{-1}. The product-containing fraction was desalted on a *NAP-10*® column and lyophilized.

Thermal-Stability Studies. Oligomer concentrations were determined by measuring the absorbance in pure H_2O at 260 nm at 80° and assuming the cytosine nucleoside analogues to have the same extinction coefficients in the denatured state as cytidine. The extinction coefficients used were: dA, $\varepsilon = 15000$; dT, $\varepsilon = 8500$; dG, $\varepsilon = 12500$; dC, $\varepsilon = 7500$. T_m Values were determined in a buffer containing 0.1M NaCl, 0.02M potassium phosphate (pH 7.5), 0.1 mM EDTA, with a 4 µM concentration for each strand. Melting curves were determined with a *Cary-100-Bio* spectrophotometer. Cuvettes were maintained at constant temps. by means of water circulation through the cuvette holder. The temp. of the soln. was measured with a thermistor directly immersed in one of the cuvettes. Temp. control and data acquisition were done automatically with an *IBM*-compatible computer. The samples were heated at a rate of 0.2° min^{-1}, and no difference was observed between heating and cooling melting curves. Melting temp. were determined by plotting the first derivative of the absorbance *vs*. temp. curve.

N^4-*Benzoyl-1-[3,5*-O-*(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-2*-O-*(2,3,5-tri*-O-*benzoyl-α*-D-*arabinofuranosyl)-β*-D-*ribofuranosyl]cytosine* (**2b**). To a cool soln. (0°) of 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-D-arabinofuranose (1.12 g, 2.22 mmol) in 1,2-dichloroethane (20 ml) under N_2, $SnCl_4$ (0.31 ml, 2.66 mmol) was added, and the soln. was kept at 0° for 10 min. After addition of *N*^{4}-benzoyl-1-[3,5-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-ribofuranosyl]cytosine (**1**; 0.86 g, 1.48 mmol), the resulting soln. was kept at 0° for 16 h. Then 10% aq. $NaHCO_3$ soln. (10 ml) was added, the suspension stirred at 0° for 20 min and filtered through *Hyflo Super Cel*, the org. layer separated, washed with H_2O (20 ml), dried, and evaporated, and the residue purified by CC (silica gel (50 g), CH_2Cl_2 (500 ml), then 1% MeOH/CH_2Cl_2): 1.27 g (83%) of **2b**. Foam. R_f (*B*) 0.45. ^{1}H-NMR ($CDCl_3$): 8.37 (*d*, *J*(6,5) = 7.5, H–C(6)); 8.08–7.91 (*m*, 8 H, Bz); 7.59–7.18 (*m*, 13 H, H–C(5), Bz); 6.01 (*s*, H–C(1′) Ara); 5.91 (*s*, H–C(1′) Cyd); 5.69 (*s*, H–C(2′) Ara); 5.56 (*d*, *J*(3′,4′) = 4.7, H–C(3′) Ara); 4.76 (*m*, H–C(4′), H_a–C(5′) Ara); 4.68 (*dd*, *J*(5′b,4′) = 4.4, *J*(5′b,5′a) = –11.8, H_b–C(5′) Ara); 4.42 (*d*, *J*(2′,3′) = 4.5, H–C(2′) Cyd); 4.33 (*d*, *J*(5′a,5′b) = –13.7, H_a–C(5′) Cyd); 4.29 (*dd*, *J*(4′,3′) = 9.5, *J*(4′,5′b) = 1.5, H–C(4′) Cyd); 4.20 (*dd*, H–C(3′) Cyd); 3.97 (*dd*, H_b–C(5′) Cyd); 1.12–0.80 (*m*, 28 H, ^{i}Pr). ^{13}C-NMR ($CDCl_3$): 166.18 (C(4)); 165.70, 165.44, 162.51 (C=O); 155.11 (C(2)); 144.30 (C(6)); 133.38, 133.34, 133.19, 132.88, 130.06, 129.94, 129.36, 129.17, 128.99, 128.32, 127.68 (Bz); 104.30 (C(1′) Ara); 96.17 (C(5)); 90.37 (C(1′) Cyd); 82.12 (C(4′) Cyd); 81.88 (C(2′) Ara); 81.13 (C(4′) Ara); 78.33 (C(2′) Cyd); 78.23 (C(3′) Ara); 66.78 (C(3′) Cyd); 63.57 (C(5′) Ara); 59.28 (C(5′) Cyd), 17.44, 17.27, 16.98, 16.81, 16.62, 13.38, 13.06, 12.87, 12.49 (^{i}Pr).

N^4-*Benzoyl-1-[3,5*-O-*(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-2*-O-*(2,3,5-tri*-O-*benzoyl-α*-L-*arabinofuranosyl)-β*-D-*ribofuranosyl]cytosine* (**2c**). As described for **2b**, with 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-L-arabinofuranose (1.08 g, 2.14 mmol), $SnCl_4$ (0.3 ml, 2.57 mmol), and **1** (844 mg, 1.43 mmol): 1.06 g (72%) of **2c**. Foam. R_f (*B*) 0.48. ^{1}H-NMR ($CDCl_3$): 8.34 (*d*, *J*(6,5) = 7.5, H–C(6)); 8.12–7.92 (*m*, 8 H, Bz); 7.61–7.23 (*m*, 13 H, H–C(5), Bz); 5.91 (*s*, H–C(1′) Cyd); 5.81 (*s*, H–C(1′) Ara); 5.68 (*dd*, *J*(3′,2′) = 1.1, *J*(3′,4′) = 4.2, H–C(3′) Ara); 5.63 (*d*, H–C(2′) Ara); 5.09 (*ddd*, *J*(4′,5′a) = 3.4, *J*(4′,5′b) = 3.7, H–C(4′) Ara); 4.84 (*dd*, *J*(5′a,5′b) = –11.8, H_a–C(5′) Ara); 4.72 (*dd*, H_b–C(5′) Ara); 4.57 (*d*, *J*(2′,3′) = 3.7, H–C(2′) Cyd); 4.32 (*m*, H–C(3′), H–C(4′), H_a–C(5′) Cyd); 4.03 (*dd*, *J*(5′b,4′) = 2.2, *J*(5′b,5′a) = –13.4, H_b–C(5′) Cyd); 1.10–0.93 (*m*, 28 H,

^{i}Pr). ^{13}C-NMR ($CDCl_3$): 166.18 (C(4)); 165.82, 165.22, 162.43 (C=O); 154.98 (C(2)); 144.21 (C(6)); 133.31, 133.29, 133.19, 132.78, 129.99, 129.82, 129.01, 128.44, 128.42, 128.20, 127.68 (Bz); 104.69 (C(1′) Ara); 96.23 (C(5)); 90.47 (C(1′) Cyd); 82.50 (C(4′) Cyd); 81.94 (C(4′) Ara); 80.86 (C(2′) Ara); 77.77 (C(2′) Cyd); 75.64 (C(3′) Ara); 69.03 (C(3′) Cyd); 63.54 (C(5′) Ara); 59.34 (C(5′) Cyd); 17.44, 17.40, 17.28, 17.06, 16.92, 16.82, 16.74, 13.36, 13.01, 12.90, 12.67 (^{i}Pr).

N^4-*Benzoyl-1-[3,5*-O-*(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-2*-O-*(2,3,4-tri*-O-*acetyl-β*-D-*ribopyranosyl)-β*-D-*ribofuranosyl]cytosine* (**2d**). As described for **2b**, with 1,2,3,4-tetra-*O*-acetyl-β-D-ribopyranose (706 mg, 2.22 mmol), $SnCl_4$ (0.31 ml, 2.66 mmol), and **1** (873 mg, 1.48 mmol) for 3 days at 0°: 1.08 g (86%) of **2d**. Foam. R_f (*B*) 0.35. ^{1}H-NMR ($CDCl_3$): 8.98 (br. *s*, NH); 8.32 (*d*, *J*(6,5) = 7.6, H–C(6)); 7.90 (*d*, *J* = 7.3, 2 H, Bz); 7.63–7.49 (*m*, 4 H, H–C(5), Bz); 5.89 (*s*, H–C(1′) Cyd); 5.44 (*d*, *J*(1′,2′) = 2.3, H–C(1′) Rib); 5.42 (*dd*, *J*(3′,2′) = 3.8, *J*(3′,4′) = 3.6, H–C(3′) Rib); 5.27 (*ddd*, H–C(4′) Rib); 5.23 (*dd*, H–C(2′) Rib); 4.56 (*d*, *J*(5′a,4′) = 2.0, *J*(5′a,5′b) = –12.7, H_a–C(5′) Rib); 4.37 (*m*, H–C(2′) Cyd); 4.30 (*d*, *J*(5′a,5′b) = –13.5, H_a–C(5′) Cyd); 4.22 (*m*, H–C(3′), H–C(4′) Cyd); 3.99 (*d*, H_b–C(5′) Cyd); 3.87 (*dd*, *J*(5′b,4′) = 2.5, H_b–C(5′) Rib); 2.13 (*s*, 1 Ac); 2.10 (*s*, 1 Ac); 2.01 (*s*, 1 Ac); 1.09–0.91 (*m*, 28 H, ^{i}Pr).

N^4-*Benzoyl-1-{2*-O-*[2,3-di*-O-*acetyl-3,5*-O-*(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β*-D-*erythrofurano-syl]-β*-D-*ribofuranosyl}cytosine* (**2e**). As described for **2b**, with 1,2,3-tri-*O*-acetyl-D-erythrose (556 mg, 2.26 mmol), $SnCl_4$ (0.32 ml, 2.71 mmol), and **1** (1.33 g, 2.26 mmol): 894 mg (51%) of **2e**. Foam. R_f (*B*) 0.32. ^{1}H-NMR ($CDCl_3$): 9.01 (br. *s*, NH); 8.29 (*d*, *J*(6,5) = 7.5, H–C(6)); 7.91 (*d*, *J* = 7.4, 2 H, Bz); 7.60–7.47 (*m*, 4 H, H–C(5), Bz); 5.78 (*s*, H–C(1′) Cyd); 5.59 (*s*, H–C(1′) Ery); 5.54 (*ddd*, *J*(3′,2′) = 5.7, *J*(3′,4′a) = 6.3, *J*(3′,4′b) = 4.0, H–C(3′) Ery); 5.33 (*d*, H–C(2′) Ery); 4.53 (*dd*, *J*(4′a,4′b) = –10.1, H_a–C(4′) Ery); 4.35 (*m*, H–C(2′) Cyd); 4.28 (*d*, *J*(5′a,5′b) = –13.5, H_a–C(5′) Cyd); 4.19 (*m*, H–C(3′), H–C(4′) Cyd); 3.98 (*d*, H_b–C(5′) Cyd); 3.94 (*dd*, H_b–C(4′) Ery); 2.05 (*s*, 1 Ac); 2.04 (*s*, 1 Ac); 1.09–0.97 (*m*, 28 H, ^{i}Pr).

N^4-*Benzoyl-1-[2*-O-*(2,3,5-tri*-O-*benzoyl-α*-D-*arabinofuranosyl)-β*-D-*ribofuranosyl]cytosine* (**3b**). To a soln. of **2b** (1.3 g, 1.26 mmol) in THF (4 ml), a soln. of $Bu_4NF \cdot 3\,H_2O$ (1.11 g, 3.53 mmol) in THF (3.5 ml) was added and kept for 15 min at 20°. The mixture was evaporated and co-evaporated with $CHCl_3$ (2 × 20 ml) and the residue applied to CC (silica gel (50 g), *A* (300 ml), then *B*): 780 mg (78%) of **3b**. Foam. R_f (*C*) 0.35. ^{1}H-NMR ($CDCl_3$): 8.39 (*d*, *J*(6,5) = 7.5, H–C(6)); 8.04–7.85 (*m*, 8 H, Bz); 7.59–7.23 (*m*, 13 H, H–C(5), Bz); 5.93 (*s*, H–C(1′) Ara); 5.92 (*d*, *J*(1′,2′) = 1.6, H–C(1′) Cyd); 5.69 (*d*, *J*(2′,3′) = 1.1, H–C(2′) Ara); 5.61 (*dd*, *J*(3′,4′) = 3.3, H–C(3′) Ara); 4.72 (*m*, H–C(4′), H_a–C(5′) Ara); 4.65 (*m*, H_b–C(5′) Ara, H–C(2′) Cyd); 4.40 (*dd*, *J*(3′,2′) = 5.6, *J*(3′,4′) = 7.3, H–C(3′) Cyd); 4.08 (*m*, H–C(4′), H_a–C(5′) Cyd); 3.87 (*dd*, *J*(5′b,4′) = 1.5, *J*(5′b,5′a) = –11.6, H_b–C(5′) Cyd). ^{13}C-NMR ($CDCl_3$): 166.19 (C(4)); 165.52, 165.40, 162.76 (C=O); 154.96 (C(2)); 145.67 (C(6)); 133.66, 133.57, 133.05, 132.88, 129.94, 129.73, 129.36, 128.86, 128.28, 128.14, 127.77 (Bz); 104.90 (C(1′) Ara); 96.88 (C(5)); 91.10 (C(1′) Cyd); 84.77 (C(4′) Cyd); 81.90 (C(2′) Ara); 81.41 (C(4′) Ara); 78.49 (C(2′) Cyd); 77.81 (C(3′) Ara); 67.82 (C(3′) Cyd); 63.62 (C(5′) Ara); 60.20 (C(5′) Cyd). LSI-MS: 792.2428 ($[C_{42}H_{37}N_3O_{13}+H]^+$; calc. 792.2404).

N^4-*Benzoyl-1-[2*-O-*(2,3,5-tri*-O-*benzoyl-α*-L-*arabinofuranosyl)-β*-D-*ribofuranosyl]cytosine* (**3c**). As described for **3b**, with **2c** (826 mg, 0.8 mmol): 500 mg (79%) of **3c**. Foam. R_f (*C*) 0.34. ^{1}H-NMR ($CDCl_3$): 9.23 (br. *s*, NH); 8.26 (*d*, *J*(6,5) = 7.5, H–C(6)); 8.05–7.83 *m* (8 H, Bz); 7.57–7.21 (*m*, 13 H, H–C(5), Bz); 5.99 (*d*, *J*(1′,2′) = 3.1, H–C(1′) Cyd); 5.67 (*dd*, *J*(3′,2′) = 1.9, *J*(3′,4′) = 5.3, H–C(3′) Ara); 5.57 (*s*, H–C(1′) Ara); 5.55 (*d*, H–C(2′) Ara); 4.77 (*m*, H–C(4′), H_a–C(5′) Ara); 4.71 (*dd*, *J*(2′,3′) = 5.0, H–C(2′) Cyd); 4.63 (*dd*, *J*(5′b,4′) = 4.9, *J*(5′b,5′a) = –13.1, H_b–C(5′) Ara); 4.51 (*dd*, *J*(3′,4′) = 5.9, H–C(3′) Cyd); 4.20 (*d*, H–C(4′) Cyd); 4.02 (*d*, *J*(5′b,5′a) = –11.5, H_a–C(5′) Cyd); 3.88 (*d*, H_b–C(5′) Cyd). ^{13}C-NMR ($CDCl_3$): 166.12 (C(4)); 166.05, 165.67, 162.61 (C=O); 154.95 (C(2)); 146.46 (C(6)); 133.65, 133.60, 133.11, 132.99, 129.94, 129.89, 129.66, 128.99, 128.87, 128.54, 128.28, 127.73 (Bz); 105.92 (C(1′) Ara); 97.02 (C(5)); 92.12 (C(1′) Cyd); 85.09 (C(4′) Cyd); 82.92 (C(4′) Ara); 80.77 (C(2′) Ara); 78.73 (C(2′) Cyd); 77.21 (C(3′) Ara); 69.23 (C(3′) Cyd); 63.26 (C(5′) Ara); 60.96 (C(5′) Cyd). LSI-MS: 792.2401 ($[C_{42}H_{37}N_3O_{13}+H]^+$; calc. 792.2404).

N^4-*Benzoyl-1-[2*-O-*(2,3,4-tri*-O-*acetyl-β*-D-*ribopyranosyl)-β*-D-*ribofuranosyl]cytosine* (**3d**). As described for **3b**, with **2d** (1.0 g, 1.18 mmol): 522 mg (73%) of **3d**. Foam. R_f (*C*) 0.28. ^{1}H-NMR ($CDCl_3$): 9.20 (br. *s*, NH); 8.52 (*d*, *J*(6,5) = 7.6, H–C(6)); 8.02 (*d*, *J* = 7.3, 2 H, Bz); 7.63–7.49 (*m*, 4 H, H–C(5), Bz); 5.87 (*d*, *J*(1′,2′) = 2.2, H–C(1′) Cyd); 5.46 (*t*, *J*(3′,2′) = *J*(3′,4′) = 3.1, H–C(3′) Rib); 5.17 (*d*, *J*(1′,2′) = 4.9, H–C(1′) Rib); 5.04 (*m*, H–C(2′), H–C(4′) Rib); 4.61 (*dd*, *J*(2′,3′) = 5.1, H–C(2′) Cyd); 4.43 (*dd*, *J*(3′,4′) = 6.4, H–C(3′) Cyd); 4.15 (*d*, H–C(4′) Cyd); 4.02 (*m*, H_a–C(5′) Cyd, H_a–C(5′) Rib); 3.87 (*d*, *J*(5′b,5′a) = –12.2, H_b–C(5′) Cyd); 3.76 (*dd*, *J*(5′b,4′) = 6.2, *J*(5′b,5′a) = –12.5, H_b–C(5′) Rib); 2.06 (*s*, Ac); 2.05 (*s*, 1 Ac); 2.04 (*s*, 1 Ac). LSI-MS: 606.1949 ($[C_{27}H_{31}N_3O_{13}+H]^+$; calc. 606.1935).

N^4-*Benzoyl-1-[2*-O-*(2,3-di*-O-*acetyl-β*-D-*erythrofuranosyl)-β*-D-*ribofuranosyl]cytosine* (**3e**). As described for **3b**, with **2e** (854 mg, 1.1 mmol): 410 mg (70%) of **3e**. Foam. R_f (*C*) 0.26. ^{1}H-NMR ($CDCl_3$): 9.06 (br. *s*, NH);

8.29 (*d*, *J*(6,5) = 7.6, H–C(6)); 7.95 (*d*, *J* = 7.3, 2 H, Bz); 7.62–7.47 (*m*, 4 H, H–C(5), Bz); 5.82 (*d*, *J*(1′,2′) = 3.4, H–C(1′) Cyd); 5.42 (*ddd*, *J*(3′,2′) = 5.6, *J*(3′,4′a) = 5.3, *J*(3′,4′b) = 3.4, H–C(3′) Ery); 5.32 (*d*, *J*(1′,2′) = 1.6, H–C(1′) Ery); 5.24 (*dd*, H–C(2′) Ery); 4.63 (*dd*, *J*(2′,3′) = 5.3, H–C(2′) Cyd); 4.43 (*dd*, *J*(3′,4′) = 5.3, H–C(3′) Cyd); 4.20 (*dd*, *J*(4′,a,4′b) = –10.3, H_a–C(4′) Ery); 4.15 (*d*, H–C(4′) Cyd); 4.01 (*d*, *J*(5′a,5′b) = –12.5, H_a–C(5′) Cyd); 3.86 (*m*, H_b–C(5′) Cyd, H_b–C(4′) Ery); 2.06 (*s*, 1 Ac); 2.05 (*s*, 1 Ac). LSI-MS: 534.1739 ($[C_{24}H_{27}N_3O_{11}+H]^+$; calc. 534.1724).

N^4-*Benzoyl-1-[5*-O-*(dimethoxytrityl)-2*-O-*(2,3,5-tri*-O-*benzoyl-α*-D-*arabinofuranosyl)-β*-D-*ribofuranosyl]cytosine* (**4b**). Following co-evaporation with anh. pyridine, **3b** (445 mg, 0.56 mmol) was dissolved in pyridine (20 ml), and dimethoxytrityl chloride ($(MeO)_2TrCl$; 285 mg, 0.84 mmol) was added. The mixture was stirred for 2 h at r.t. (TLC monitoring: reaction completed). Following neutralization with 10% aq. $NaHCO_3$ soln., the mixture was concentrated and partitioned twice between CH_2Cl_2 and aq. $NaHCO_3$ soln. The org. layer was evaporated and the residue purified by CC (silica gel (25 g), 0 → 1% MeOH/CH_2Cl_2 containing 0.5% of pyridine): 573 mg (93%) of **4b**. Foam. ^{1}H-NMR ($CDCl_3$): 8.69 (br. *s*, NH); 8.49 (*d*, *J*(6,5) = 7.3, H–C(6)); 8.04–7.87 (*m*, 8 H, Bz); 7.61–7.13 (*m*, 22 H, H–C(5), Bz, Ph); 6.88 (*d*, *J* = 9.0, 4 H, Tr); 6.09 (*s*, H–C(1′) Cyd); 6.07 (*s*, H–C(1′) Ara); 5.72 (*d*, *J*(2′,3′) = 1.3, H–C(2′) Ara); 5.61 (*dd*, *J*(3′,4′) = 2.4, H–C(3′) Ara); 4.72 (*m*, H–C(4′), H_a–C(5′), H_b–C(5′) Ara); 4.54 (*d*, *J*(2′,3′) = 5.8, H–C(2′) Cyd); 4.50 (*ddd*, *J*(3′,4′) = 8.3, H–C(3′) Cyd); 4.12 (*ddd*, *J*(4′,5′a) = 1.9, *J*(4′,5′b) = 2.9, H–C(4′) Cyd); 3.82 (*s*, 1 MeO); 3.81 (*s*, 1 MeO); 3.66 (*dd*, *J*(5′a,5′b) = –11.2, H_a–C(5′) Cyd); 3.54 (*dd*, H_b–C(5′) Cyd); 2.69 (*d*, *J* = 9.3, OH). ^{13}C-NMR ($CDCl_3$): 166.15 (C(4)); 165.44, 165.27, 162.41 (C=O); 158.75 (Tr); 155.02 (C(2)); 144.51 (C(6)); 144.14 (Tr); 135.62, 135.35, 133.65, 133.55, 133.01, 130.12, 130.03, 129.77, 129.62, 128.97, 128.86, 128.50, 128.26, 128.04, 127.53, 127.16 (Bz, Tr); 113.36 (Tr); 104.74 (C(1′) Ara); 96.55 (C(5)); 89.73 (C(1′) Cyd); 87.09 ($ArPh_2C$); 83.38 (C(4′) Cyd); 82.11 (C(2′) Ara); 81.18 (C(4′) Ara); 78.70 (C(2′) Cyd); 78.06 (C(3′) Ara); 67.89 (C(3′) Cyd); 63.74 (C(5′) Ara); 60.95 (C(5′) Cyd); 55.21 (MeO). ESI-MS (pos.): 1094.3716 ($[C_{63}H_{55}N_3O_{15}+H]^+$; calc. 1094.3711). LSI-MS (pos.; ThGly/NaOAc 1116 ($[M+Na]^+$).

N^4-*Benzoyl-1-[5*-O-*(dimethoxytrityl)-2*-O-*(2,3,5-tri*-O-*benzoyl-α*-L-*arabinofuranosyl)-β*-D-*ribofuranosyl]cytosine* (**4c**). As described for **4b**, with **3c** (265 mg, 0.33 mmol), $(MeO)_2TrCl$ (170 mg, 0.50 mmol), and pyridine (20 ml), followed by CC (silica gel (25 g), 0 → 1% MeOH/CH_2Cl_2 containing 0.5% of pyridine): 350 mg (95%) of **4c**. Foam. ^{1}H-NMR ($CDCl_3$): 8.60 (br. *s*, NH); 8.40 (*d*, *J*(6,5) = 7.4, H–C(6)); 8.08–7.88 (*m*, 8 H, Bz); 7.68–7.25 (*m*, 22 H, H–C(5), Bz, Tr); 6.87 (*d*, *J* = 8.8, 4 H, Tr); 6.25 (*s*, H–C(1′) Cyd); 5.75 (*dd*, *J*(3′,2′) = 2.4, *J*(3′,4′) = 5.9, H–C(3′) Ara); 5.64 (*s*, H–C(1′) Ara); 5.52 (*d*, H–C(2′) Ara); 4.95 (*ddd*, *J*(4′,5′a) = 3.4, *J*(4′,5′b) = 3.9, H–C(4′) Ara); 4.84 (*dd*, *J*(5′a,5′b) = –12.2, H_a–C(5′) Ara); 4.68 (*dd*, H_b–C(5′) Ara); 4.51 (*m*, H–C(2′), H–C(3′) Cyd); 4.22 (*ddd*, *J*(4′,3′) = 6.8, *J*(4′,5′a) = 2.2, *J*(4′,5′b) = 2.9, H–C(4′) Cyd); 3.81 (*s*, 2 MeO); 3.64 (*dd*, *J*(5′a,5′b) = –11.2, H_a–C(5′) Cyd); 3.56 (*dd*, H_b–C(5′) Cyd); 2.99 (br. *s*, OH). ^{13}C-NMR ($CDCl_3$): 166.28 (C(4)); 166.06, 165.68, 162.20 (C=O); 158.72 (Tr); 157.47 (C(2)); 144.57 (C(6)); 144.12, 135.90, 135.53, 135.31, 133.65, 133.51, 133.12, 132.88, 130.11, 129.98, 129.91, 129.76, 128.93, 128.73, 128.48, 128.23, 127.56, 127.14 (Bz, Tr); 113.35 (Tr); 105.84 (C(1′) Ara); 96.66 (C(5)); 89.50 (C(1′) Cyd), 87.14 ($ArPh_2C$); 83.36, 83.25 (C(4′)); 80.52, 79.45 (C(2′)); 77.00 (C(3′) Ara); 69.28 (C(3′) Cyd); 63.17 (C(5′) Ara); 61.50 (C(5′) Cyd); 55.19 (MeO). ESI-MS (pos.): 1094.3707 ($[C_{63}H_{55}N_3O_{15}+H]^+$; calc. 1094.3711).

N^4-*Benzoyl-1-[5*-O-*(dimethoxytrityl)-2*-O-*(2,3,4-tri*-O-*acetyl-β*-D-*ribopyranosyl)-β*-D-*ribofuranosyl]cytosine* (**4d**). As described for **4b**, with **3d** (339 mg, 0.56 mmol), $(MeO)_2TrCl$ (285 mg, 0.84 mmol), and pyridine (20 ml), followed by CC (silica gel (25 g), 0 → 1% MeOH/CH_2Cl_2 containing 0.5% of pyridine): 488 mg (94%) of **4d**. Foam. ^{1}H-NMR ($CDCl_3$): 8.75 (br. *s*, NH); 8.52 (*d*, *J*(6,5) = 7.3, H–C(6)); 7.90 (*d*, *J* = 7.3, 2 H, Bz); 7.62–7.16 (*m*, 13 H, H–C(5), Bz, Tr); 6.87 (*d*, *J* = 8.8, 4 H, Tr); 6.18 (*s*, H–C(1′) Cyd); 5.53 (*dd*, *J*(3′,2′) = 3.4, *J*(3′,4′) = 3.0, H–C(3′) Rib); 5.38 (*d*, *J*(1′,2′) = 5.4, H–C(1′) Rib); 5.11 (*ddd*, *J*(4′,5′a) = 4.1, *J*(4′,5′b) = 6.9, H–C(4′) Rib); 5.07 (*dd*, H–C(2′) Rib); 4.45 (*dd*, *J*(3′,2′) = 4.9, *J*(3′,4′) = 7.3, H–C(3′) Cyd); 4.38 (*d*, H–C(2′) Cyd); 4.15 (*m*, H–C(4′) Cyd, H_a–C(5′) Rib); 3.93 (*dd*, *J*(5′b,5′a) = –12.2, H_b–C(5′) Rib); 3.82 (*s*, 2 MeO); 3.65 (*dd*, *J*(5′a,4′) = 2.0, *J*(5′a,5′b) = –11.2, H_a–C(5′) Cyd); 3.54 (*dd*, *J*(5′ b,4′) = 2.9, H_b–C(5′) Cyd); 2.10 (*s*, 1 Ac); 2.06 (*s*, 1 Ac), 2.04 (*s*, 1 Ac). ^{13}C-NMR ($CDCl_3$): 170.06, 169.87, 169.75 (C=O); 166.20 (C(4)); 162.43 (C=O); 158.73 (Tr); 154.96 (C(2)); 144.94 (C(6)); 135.59, 135.34, 133.08, 130.10, 130.03, 128.97, 128.27, 128.18, 127.55, 127.16 (Bz, Tr); 113.36 (Tr); 99.44 (C(1′) Rib); 96.44 (C(5)); 89.46 (C(1′) Cyd); 87.16 ($ArPh_2C$); 82.77, 81.89 (C(4′), C(2′)); 68.56, 68.43, 67.02, 66.55 (C(3′) Cyd, C(2′), C(3′), C(4′) Rib); 61.94 (C(5′) Rib); 60.87 (C(5′) Cyd); 55.20 (MeO); 20.69 (Ac). ESI-MS (pos.): 908.3247 ($[C_{48}H_{49}N_3O_{15}+H]^+$; calc. 908.3241). LSI-MS (pos; ThGly/NaOAc): 930 ($[M+Na]^+$).

N^4-*Benzoyl-1-[2*-O-*(2,3-di*-O-*acetyl-β*-D-*erythrofuranosyl)-5*-O-*(dimethoxytrityl)-β*-D-*ribofuranosyl]cytosine* (**4e**). As described for **4b**, with **3e** (244 mg, 0.46 mmol), $(MeO)_2TrCl$ (235 mg, 0.69 mmol), and pyridine (20 ml), followed by CC (silica gel (25 g), 0 → 1% MeOH/CH_2Cl_2 containing 0.5% of pyridine: 336 mg (87%) of

4e. Foam. ^{1}H-NMR ($CDCl_3$): 8.93 (br. *s*, NH); 8.41 (*d*, *J*(6,5) = 7.3, H–C(6)); 7.91 (*d*, *J* = 7.3, 2 H, Bz); 7.69–7.24 (*m*, 13 H, H–C(5), Bz, Tr); 6.87 (*d*, *J* = 9, 4 H, Tr); 6.14 (*d*, *J*(1′,2′) = 1.9, H–C(1′) Cyd); 5.51 (*d*, *J*(1′,2′) = 1.5, H–C(1′) Ery); 5.47 (*ddd*, *J*(3′,2′) = 5.4, *J*(3′,4′a) = 5.4, *J*(3′,4′b) = 3.4, H–C(3′) Ery); 5.27 (*dd*, H–C(2′) Ery); 4.46 (*dd*, *J*(3′,2′) = 4.9, *J*(3′,4′) = 7.8, H–C(3′) Cyd); 4.39 (*dd*, H–C(2′) Cyd); 4.36 (*dd*, *J*(4′a,4′b) = –10.2, H_a–C(4′) Ery); 4.16 (*ddd*, *J*(4′,5′a) = 2.4, *J*(4′,5′b) = 2.9, H–C(4′) Cyd); 3.96 (*dd*, H_b–C(4′) Ery); 3.82 (*s*, 2 MeO); 3.61 (*dd*, *J*(5′a,5′b) = –11.2, H_a–C(5′) Cyd); 3.53 (*dd*, H_b–C(5′) Cyd); 3.18 (br. *s*, OH); 2.08 (*s*, 1 Ac); 2.06 (*s*, 1 Ac). ^{13}C-NMR ($CDCl_3$): 170.14, 170.01 (C=O); 166.23 (C(4)); 162.33 (C=O); 158.70 (Tr); 154.12 (C(2)); 144.79 (C(6)); 135.89, 135.58, 132.99, 130.10, 130.03, 128.92, 128.25, 128.00, 127.62, 127.12 (Bz, Tr); 113.33 (Tr); 106.20 (C(1′) Ery); 96.94 (C(5)); 89.33 (C(1′) Cyd); 87.10 (ArPh_2*C*); 83.09 (C(4′) Cyd); 80.37 (C(2′) Cyd); 76.42 (C(4′) Ery); 71.26, 70.16 (C(2′), C(3′) Ery); 68.79 (C(3′) Cyd); 61.43 (C(5′)); 55.19 (MeO); 20.50 (Ac). ESI-MS (pos.): 836.3041 ($[C_{45}H_{45}N_3O_{13}+H]^+$; calc. 836.3030).

N^4-*Benzoyl-1-[5*-O-*(dimethoxytrityl)-2*-O-*(2,3,5-tri*-O-*benzoyl-α*-D-*arabinofuranosyl)-β*-D-*ribofuranosyl]-cytosine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**5b**). Derivative **4b** (545 mg, 0.5 mmol) was dissolved in CH_2Cl_2 (6 ml) under Ar and ^{i}Pr$_2$EtN (270 µl, 1.5 mmol) and 2-cyanoethyl diisopropylphosphoramidochloridite (280 µl, 1.25 mmol) were added. The soln. was stirred for 75 min (TLC: complete reaction). H_2O (4 ml) was added, the soln. stirred for 10 min and partitioned between CH_2Cl_2 (50 ml) and aq. $NaHCO_3$ soln. (30 ml), and the org. layer washed with aq. NaCl soln. (3 × 30 ml). The aq. phases were back extracted with CH_2Cl_2 (20 ml). Evaporation of the org. phase left an oil, which was purified by FC (silica gel (35 g), hexane/AcOEt/Et_3N 42:56:2) to give **5b** as a foam, after co-evaporation with CH_2Cl_2. Dissolution in CH_2Cl_2 (2 ml) and precipitation in cold (–70°) hexane (100 ml) afforded **5b** (587 mg, 90%). White powder. R_f (hexane/AcOEt/Et_3N 42:56:2) 0.30. ^{31}P-NMR ($CDCl_3$): 150.33; 151.87. ESI-MS (pos.): 1294.4762 ($[C_{72}H_{72}N_5O_{16}P+H]^+$; calc. 1294.4789).

N^4-*Benzoyl-1-[5*-O-*(dimethoxytrityl)-2*-O-*(2,3,5-tri*-O-*benzoyl-α*-L-*arabinofuranosyl)-β*-D-*ribofuranosyl]-cytosine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**5c**). As described for **5b**, with **4c** (325 mg, 0.30 mmol), ^{i}Pr$_2$EtN (160 µl, 0.9 mmol), and 2-cyanoethyl diisopropylphosphoramidochloridite (170 µl, 0.75 mmol). FC (silica gel (40 g), hexane/acetone/Et_3N 68:30:2) gave **5c** as a foam, after co-evaporation with CH_2Cl_2. Dissolution in CH_2Cl_2 (2 ml) and precipitation in cold (–70°) hexane (100 ml) afforded **5c** (321 mg, 82%). White powder. R_f (hexane/acetone/Et_3N 49:49:2) 0.55. ^{31}P-NMR ($CDCl_3$): 149.87; 151.94. ESI-MS (pos.): 1316.4585 ($[C_{72}H_{72}N_5O_{16}P+Na]^+$; calc. 1316.4609).

N^4-*Benzoyl-1-[5*-O-*(dimethoxytrityl)-2*-O-*(2,3,4-tri*-O-*acetyl-β*-D-*ribopyranosyl)-β*-D-*ribofuranosyl]cytosine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**5d**). As described for **5b**, with **4d** (508 mg, 0.55 mmol), ^{i}Pr$_2$EtN (290 µl, 1.65 mmol), and 2-cyanoethyl diisopropylphosphoramidochloridite (315 µl, 1.4 mmol). FC (silica gel (40 g), hexane/acetone/Et_3N 62:36:2) gave **5d** as a foam, after co-evaporation with CH_2Cl_2. Dissolution in CH_2Cl_2 (2 ml) and precipitation in cold (–70°) hexane (100 ml) afforded **5d** (485 mg, 79%). White powder. R_f (hexane/acetone/Et_3N 49:49:2) 0.40. ^{31}P-NMR ($CDCl_3$): 149.77; 152.67. ESI-MS (pos.): 1108.4316 ($[C_{57}H_{66}N_5O_{16}P+H]^+$; calc. 1108.4320).

N^4-*Benzoyl-1-[2*-O-*(2,3-di*-O-*acetyl-β*-D-*erythrofuranosyl)-5*-O-*(dimethoxytrityl)-β*-D-*ribofuranosyl]cytosine 3′-(2-Cyanoethyl Diisopropylphosphoramidite)* (**5e**). As described for **5b**, with **4e** (334 mg, 0.40 mmol), ^{i}Pr$_2$EtN (210 µl, 1.20 mmol), and 2-cyanoethyl diisopropylphosphoramidochloridite (225 µl, 1.0 mmol). FC (silica gel (30 g), hexane/acetone/Et_3N 60:38:2) gave **5e** as a foam after co-evaporation with CH_2Cl_2. Dissolution in CH_2Cl_2 (2 ml) and precipitation in cold (–70°) hexane (60 ml) afforded **5d** (405 mg, 97%). White powder. R_f (hexane/acetone/Et_3N 49:49:2) 0.40. ^{31}P-NMR ($CDCl_3$): 150.33; 151.87. ESI-MS (pos.): 1036.4102 ($[C_{54}H_{62}N_5O_{14}P+H]^+$; calc. 1036.4109).

1,2,3-Tri-O-*acetyl-5-azido-5-deoxy*-D-*ribofuranose* (**7**). Tosyl chloride (1.87 g, 9.8 mmol) was added to a soln. of 1,2,3-tri-*O*-acetyl-D-ribofuranose (**6**) [21] (1.93 g, 7.0 mmol) in pyridine/1,2-dichloroethane 1:4 (25 ml). The mixture was kept for 16 h at 20°, after which MeOH (1 ml) was added. After stirring for 30 min at 20°, the mixture was diluted with CH_2Cl_2, the org. layer washed with 10% aq. $NaHCO_3$ soln. (30 ml) and H_2O (30 ml), dried (Na_2SO_4), and evaporated, and the residue co-evaporated with toluene (2 × 10 ml). The residue was dissolved in DMF (18 ml), and NaN_3 (0.7 g, 10.77 mmol) was added. The mixture was heated under stirring at 80° for 2 h and, after cooling to r.t., diluted with Et_2O (100 ml) and washed with H_2O (3 × 30 ml). The org. layer was dried (Na_2SO_4) and evaporated and the residue applied to CC (silica gel (100 g), *A*, then 0.2% MeOH/CH_2Cl_2): **7** (1.62 g, 77%). Oil. R_f (*E*) 0.50 (*β*-D-isomer), 0.42 (*α*-D-isomer). ^{1}H-NMR ($CDCl_3$): 6.44 (*d*, *J*(1,2) = 4.3, 0.5 H, H–C(1)*α*); 6.16 (*s*, 0.5 H, H–C(1)*β*); 5.40 (*dd*, *J*(3,2) = 4.8, *J*(3,4) = 7.4, 0.5 H, H–C(3)*β*); 5.35 (*d*, 0.5 H, H–C(2)*β*); 5.20 (*dd*, *J*(2,3) = 6.9, 0.5 H, H–C(2)*α*); 5.16 (*dd*, *J*(3,4) = 3.0, 0.5 H, H–C(3)*α*); 4.34 (*ddd*, 0.5 H, H–C(4)*α*); 4.31 (*ddd*, 0.5 H, H–C(4)*β*); 3.64 (*dd*, *J*(5a,4) = 3.3, *J*(5a,5b) = –13.5, 0.5 H, H_a–C(5)*β*); 3.63 (*dd*, *J*(5a,4) = 3.3, *J*(5a,5b) = –13.2, 0.5 H, H_a–C(5)*α*); 3.53 (*dd*, *J*(5b,4) = 3.8, 0.5 H,

H_b–C(5)α); 3.27 (*dd*, *J*(5b,4) = 4.1, 0.5 H, H_b–C(5)β); 2.12, 2.11, 2.08, 2.05 (4*s*, Ac). ^{13}C-NMR ($CDCl_3$): 169.72, 169.17 (C=O); 97.97 (C(1)β); 94.11 (C(1)α); 82.89 (C(4)α); 80.34 (C(4)β); 74.21 (C(2)β); 70.29 (C(3)β); 70.20 (C(2)α); 70.05 (C(3)α); 51.84 (C(5)α); 51.35 (C(5)β); 20.85, 20.49, 20.34, 20.15 (Ac).

N^4-*Benzoyl-1-[2*-O-*(2,3-di*-O-*acetyl-5-azido-5-deoxy-*β-D-*ribofuranosyl)-3,5*-O-*(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-*β-D-*ribofuranosyl]cytosine* (**8**). As described for **2b**, with **7** (504 mg, 1.67 mmol), $SnCl_4$ (0.23 ml, 2 mmol), and **1** (820 mg, 1.39 mmol): 774 mg (67%) of **8**. Foam. R_f 0.34 (*B*), 0.75 (*C*). ^{1}H-NMR ($CDCl_3$): 8.78 (br. *s*, NH); 8.32 (*d*, *J*(6,5) = 7.7, H–C(6)); 7.89 (*d*, *J* = 7.3, 2 H, Bz); 7.62–7.48 (*m*, 4 H, H–C(5), Bz); 5.86 (*s*, H–C(1′) Cyd); 5.72 (*s*, H–C(1′) Rib); 5.41 (*m*, H–C(2′), H–C(3′) Rib); 4.41–3.97 (*m*, 7 H, H–C(2′), H–C(3′), H–C(4′), 2 H–C(5′) Cyd, H–C(4′), H_a–C(5′) Rib); 3.42 (*dd*, *J*(5′b,4′) = 4.0, *J*(5′a,5′b) = –12.8, H_b–C(5′) Rib); 2.10 (*s*, 1 Ac); 2.07 (*s*, 1 Ac); 1.12–0.98 (*m*, 28 H, ^{i}Pr). ^{13}C-NMR ($CDCl_3$): 169.78, 169.54 (C=O); 162.47 (C(4)); 154.50 (C(2)); 144.17 (C(6)); 133.18, 129.05, 127.53 (Bz); 104.35 (C(1′) Rib); 96.18 (C(5)); 89.96 (C(1′) Cyd); 81.61; 80.55 (C(4′)); 76.55; 74.57 (C(2′)); 72.72; 68.90 (C(3′)); 59.18 (C(5′) Cyd); 53.66 (C(5′) Rib); 20.37 (Ac); 17.30, 17.15, 16.94, 16.70, 16.63, 13.20, 12.90, 12.72, 12.42 (^{i}Pr). ESI-MS (pos.): 831.3409 ($[C_{37}H_{54}N_6O_{12}Si_2 + H]^+$; calc. 831.3417).

N^4-*Benzoyl-1-[2*-O-*(2,3-di*-O-*acetyl-5-azido-5-deoxy-*β-D-*ribofuranosyl)-*β-D-*ribofuranosyl]cytosine* (**9**). To a soln. of **8** (722 mg, 0.87 mmol) in THF (2 ml), 1M $Bu_4NF \cdot 3\,H_2O$ in THF (2.5 ml) was added and kept for 15 min at 20°. The mixture was evaporated, co-evaporated with $CHCl_3$ (2 × 10 ml), and applied to CC (silica gel (30 g); *A* (300 ml), then 3% MeOH/CH_2Cl_2): 461 mg (90%) of **9**. Foam. R_f (*C*) 0.25. ^{1}H-NMR ($CDCl_3$): 9.18 (br. *s*, NH); 8.40 (*d*, *J*(6,5) = 7.8, H–C(6)); 7.90 (*d*, *J* = 7.4, 2 H, Bz); 7.61–7.49 (*m*, 4 H, H–C(5), Bz); 5.94 (*d*, *J*(1′,2′) = 2.2, H–C(1′) Cyd); 5.52 (*s*, H–C(1′) Rib); 5.36 (*d*, *J*(2′,3′) = 5.9, H–C(2′) Rib); 5.31 (*dd*, *J*(3′,4′) = 5.5, H–C(3′) Rib); 4.60 (*dd*, *J*(2′,3′) = 4.8, H–C(2′) Cyd); 4.46 (*dd*, *J*(3′,4′) = 6.2, H–C(3′) Cyd); 4.19 (*m*, H–C(4′) Cyd, H–C(4′) Rib); 4.07 (*d*, *J*(5′a,5′b) = –12.5, H_a–C(5′) Cyd); 3.91 (*d*, H_b–C(5′) Cyd); 3.59 (*dd*, *J*(5′a,4′) = 5.1, *J*(5′a,5′b) = –13.2, H_a–C(5′) Rib); 3.50 (*dd*, *J*(5′b,4′) = 4.0, H_b–C(5′) Rib); 2.08 (*s*, 1 Ac); 2.05 (*s*, 1 Ac). ^{13}C-NMR ($CDCl_3$): 170.02 (C=O); 162.89 (C(4)); 155.37 (C(2)); 146.99 (C(6)); 133.27, 132.97, 129.02, 127.78 (Bz); 105.74 (C(1′) Rib); 96.91 (C(5)); 91.96 (C(1′) Cyd); 84.62; 79.91 (C(4′)); 77.00; 74.85 (C(2′)); 71.63; 68.44 (C(3′)); 60.40 (C(5′) Cyd); 52.72 (C(5′) Rib); 20.49, 20.40 (Ac). ESI-MS (pos.): 589.1863 ($[C_{25}H_{28}N_6O_{11} + H]^+$; calc. 589.1894).

N^4-*Benzoyl-1-{2*-O-*[2,3-di*-O-*acetyl-5-deoxy-5-[(trifluoroacetyl)amino]-*β-D-*ribofuranosyl]-*β-D-*ribofuranosyl}cytosine* (**10**). A mixture of **9** (424 mg, 0.72 mmol), Bu_3SnH (0.58 ml, 2.16 mmol), and AIBN (50 mg) in dioxane (15 ml) was stirred at 90° for 40 min. Ethyl trifluoroacetate (1.29 ml, 10.8 mmol) in dioxane (1.5 ml) was added in portions by syringe during this period *via* a septum (directly to the reaction mixture). TLC (toluene/AcOEt) 1:3: product **10** moved faster (R_f 0.29) than **9** (R_f 0.14). The presence of free amine was verified by TLC *D* (R_f 0.08). After cooling to r.t., the solvent was evaporated and the residue co-evaporated with $CHCl_3$ (2 × 10 ml) and applied to CC (silica gel (30 g), *A* (300 ml), then 3% MeOH/CH_2Cl_2): 310 mg (59%) of **9**. Foam. R_f (*D*) 0.64. ^{1}H-NMR ($CDCl_3$): 9.18 (br. *s*, NH); 8.79 (*dd*, *J*(NH,5′a) = 5.8, *J*(NH,5′b) = 6.3, $NHCOCF_3$); 8.58 (*d*, *J*(6,5) = 7.3, H–C(6)); 7.88 (*d*, *J* = 7.3, 2 H, Bz); 7.59 (*m*, 2 H, H–C(5), Bz); 7.48 (*t*, *J* = 7.3, 2 H, Bz); 5.89 (*d*, *J*(1′,2′) = 1.5, H–C(1′) Cyd); 5.35 (*d*, *J*(2′,3′) = 4.9, H–C(2′) Rib); 5.33 (*s*, H–C(1′) Rib); 5.14 (*dd*, *J*(3′,4′) = 7.8, H–C(3′) Rib); 4.38 (*dd*, *J*(2′,3′) = 4.6, H–C(2′) Cyd); 4.34 (*m*, H–C(3′) Cyd, H–C(4′) Rib); 4.13 (*ddd*, *J*(4′,3′) = 8.0, H–C(4′) Cyd); 4.08 (*dd*, *J*(5′a,4′) = 2.0, *J*(5′a,5′b) = –12.5, H_a–C(5′) Cyd); 3.92 (*dd*, *J*(5′b,4′) = 1.9, H_b–C(5′) Cyd); 3.70 (*ddd*, *J*(5′a,4′) = 3.4, *J*(5′a,5′b) = –14.1, H_a–C(5′) Rib); 3.62 (*ddd*, *J*(5′b,4′) = 6.6, H_b–C(5′) Rib); 2.06 (*s*, 2 Ac). ^{13}C-NMR ($CDCl_3$): 170.36, 166.81 (C=O); 162.92 (C(4)); 158.20 (*q*, *J*(C,F) = 37.1, *CO*CF_3); 155.49 (C(2)); 145.56 (C(6)); 133.42, 132.75, 129.05, 127.69 (Bz); 116.03 (*q*, *J*(C,F) 288.1, CF_3); 105.68 (C(1′) Rib); 97.18 (C(5)); 90.23 (C(1′) Cyd); 83.53; 79.94 (C(4′)); 78.88, 74.88 (C(2′)); 71.29; 68.41 (C(3′)); 59.73 (C(5′) Cyd); 41.76 (C(5′) Rib); 20.46, 20.34 (Ac). ESI-MS (pos.): 659.1825 ($[C_{27}H_{29}F_3N_4O_{12} + H]^+$; calc. 659.1812).

N^4-*Benzoyl-1-{2*-O-*[2,3-di*-O-*acetyl-5-deoxy-5-[(trifluoroacetyl)amino]-*β-D-*ribofuranosyl}-5*-O-*(dimethoxytrityl)-*β-D-*ribofuranosyl}cytosine* (**11**). As described for **4b**, with **10** (306 mg, 0.46 mmol), $(MeO)_2TrCl$ (235 mg, 0.69 mmol), and pyridine (20 ml). Purification by CC (silica gel (25 g), 0 → 1% MeOH/CH_2Cl_2 containing 0.5% of pyridine) afforded 410 mg (93%) of **11**. Foam. ^{1}H-NMR ($CDCl_3$): 9.09 (*t*, *J*(NH,5′) = 5.5, $NHCOCF_3$); 8.78 (br. *s*, NH); 8.61 (*d*, *J*(6,5) = 7.3, H–C(6)); 7.89 (*d*, *J* = 7.2, 2 H, Bz); 7.67–7.15 (*m*, 13 H, H–C(5), Bz, Tr); 6.88 (*d*, *J* = 9.1, 4 H, Tr); 5.90 (*s*, H–C(1′) Cyd); 5.29 (*m*, H–C(1′), H–C(2′) Rib); 5.09 (*dd*, *J*(3′,2′) = 4.8 *J*(3′,4′) = 7.9, H–C(3′) Rib); 4.55–4.18 (*m*, H–C(2′), H–C(3′), H–C(4′) Cyd, H–C(4′) Rib); 3.83 (*s*, 2 MeO); 3.80–3.51 (*m*, CH_2(5′) Cyd, CH_2(5′) Rib); 2.09 (*s*, 1 Ac); 2.08 (*s*, 1 Ac). ^{13}C-NMR ($CDCl_3$): 170.39, 170.05, 166.82 (C=O); 162.74 (C(4)); 158.82 (Tr); 157.80 (*q*, *J*(C,F) = 38.1, *C*OCF_3); 155.10 (C(2)); 144.95 (C(6)); 135.49, 135.24, 133.33, 132.88, 130.11, 129.11, 128.26, 128.14, 127.50, 127.29 (Bz, Tr); 115.58 (*q*, *J*(C,F) = 288.6, CF_3); 113.39 (Tr); 105.80 (C(1′) Rib); 97.09 (C(5)); 90.05 (C(1′) Cyd); 87.29 ($ArPh_2C$), 82.43;

79.67 (C(4′)); 79.31; 75.12 (C(2′)); 70.87; 69.05 (C(3′)); 60.49 (C(5′) Cyd); 55.18 (MeO); 41.49 (C(5′) Rib); 20.49, 20.31 (Ac). ESI-MS (pos.): 961.3115 ($[C_{48}H_{47}F_3N_4O_{14}+H]^+$; calc. 961.3118).

N^4-Benzoyl-1-{2-O-{2,3-di-O-acetyl-5-deoxy-5-[(trifluoroacetyl)amino]-β-D-ribofuranosyl}-5-O-(dimethoxytrityl)-β-D-ribofuranosyl}cytosine 3′-(2-Cyanoethyl Diisopropylphosphoramidite) (**12**). As described for **5b**, with **11** (410 mg, 0.42 mmol), $^iPr_2EtN$ (225 µl, 1.28 mmol), and 2-cyanoethyl diisopropylphosphoramidochloridite (145 µl, 0.64 mmol), FC (silica gel (30 g), hexane/acetone/Et_3N 68:30:2) gave **12** as a foam, after coevaporation with CH_2Cl_2. Dissolution in CH_2Cl_2 (2 ml) and precipitation in cold (−70°) hexane (60 ml) afforded **12** (317 mg, 64%). White powder. R_f (hexane/acetone/Et_3N 49:49:2) 0.53. ^{31}P-NMR ($CDCl_3$): 149.50, 152.97. ESI-MS (pos.): 1161.4200 ($[C_{57}H_{64}F_3N_6O_{15}P+H]^+$; calc. 1161.4197).

Dr. *S. Mikhailov* is grateful to the *K. U. Leuven* for a Senior Research Fellowship. The authors thank the *K. U. Leuven RFBR*, and *NATO* for financial support (GOA 97/11) and *Chantal Biernaux* for editorial help.

REFERENCES

[1] A. De Mesmaker, R. Haner, P. Martin, H. Moser, *Acc. Chem. Res.* **1995**, *28*, 366.
[2] M. Matteuchi, in 'Antisense Therapeutics', Ed. G. L. Trainor, Escom, Leiden, 1996, p. 1.
[3] M. Manoharan, *Biochim. Biophys. Acta* **1999**, *1489*, 117.
[4] B. Cuenoud, F. Casset, D. Huskin, F. Natt, R. M. Wolf, K.-H. Altmann, P. Martin, H. Moser, *Angew. Chem., Int. Ed*. **1998**, *37*, 1288.
[5] R. H. Griffey, B. P. Monia, L. L. Cummins, S. M. Freier, M. J. Greig, C. J. Guinosso, E. A. Lesnik, S. M. Manalili, V. Mohan, S. Owens, B. R. Ross, H. Sasmor, E. Wancewicz, K. Weiler, P. D. Wheeler, P. D. Cook, *J. Med. Chem*. **1996**, *39*, 5100.
[6] T. P. Prakash, M. Manoharan, A. S. Fraser, A. M. Kawasaki, E. A. Lesnik, S. Owens, *Tetrahedron Lett*. **2000**, *41*, 4855.
[7] P. A. Limbach, P. F. Crain, J. A. McCloskey, *Nucleic Acids Res.* **1994**, *22*, 2183.
[8] S. N. Mikhailov, E. V. Efimtseva, G. V. Gurskaya, V. E. Zavodnik, A. De Bruyn, J. Rozenski, P. Herdewijn, *J. Carbohydr. Chem.* **1997**, *16*, 75.
[9] E. V. Efimtseva, B. S. Ermolinsky, M. V. Fomitcheva, S. V. Meshkov, N. Sh. Padyukova, S. N. Mikhailov, A. Van Aerschot, J. Rozenski, P. Herdewijn, *Collect. Czech. Chem. Commun. Special Issue* **1996**, *61*, 206.
[10] V. L. Tunitskaya, E. E. Rusakova, L. V. Memelova, S. N. Kochetkov, A. Van Aerschot, P. Herdewijn, E. V. Efimtseva, B. S. Ermolinsky, S. N. Mikhailov, *FEBS Letters* **1999**, *442*, 20.
[11] O. M. Gritsenko, S. N. Mikhailov, E. V. Efimtseva, A. Van Aerschot, P. Herdewijn, E. S. Gromova, *Nucleosides Nucleotides Nucleic Acids* **2000**, *19*, 1805.
[12] I. Luyten, R. M. Esnouf, S. N. Mikhailov, E. V. Efimtseva, P. Michiels, H. A. Heus, C. W. Hilbers, P. Herdewijn, *Helv. Chim. Acta* **2000**, *83*, 1278.
[13] S. Hanessian, J. Banoub, in 'Methods in Carbohydrate Chemistry', Academic Press, New York, 1980, p. 243.
[14] I. A. Korbuch, L. N. Abramova, M. N. Preobrazhenskaya, in 'Nucleic Acid Chemistry', Eds. L. B. Townsend and R. S. Tipson, Wiley-Interscience Publication, New-York, 1978, Part 1, p. 151.
[15] H. Vorbruggen, B. Bennua, *Chem. Ber.* **1981**, *114*, 1279.
[16] S. N. Mikhailov, E. De Clercq, P. Herdewijn, *Nucleosides Nucleotides* **1996**, *15*, 1323.
[17] R. Andersson, O. Theander, E. Westerlund, *Carbohydr. Res.* **1978**, *61*, 501.
[18] L. N. Beigelman, S. N. Mikhailov, *Carbohydr. Res.* **1990**, *203*, 324.
[19] J. P. Horwitz, A. J. Tomson, J. A. Urbanski, J. Chua, *J. Org. Chem*. **1962**, *27*, 3045.
[20] T.-S. Lin, J. P. Neenan, Y.-C. Cheng, W. H. Prusoff, D. C. Ward, *J. Med. Chem*. **1976**, *19*, 495.
[21] M. Bartra, F. Urpi, J. Vilarrasa, *Tetrahedron Lett*. **1987**, *28*, 5941, and refs. cit. therein.
[22] M. Bartra, P. Romea, F. Urpi, J. Vilarrasa, *Tetrahedron* **1990**, *46*, 587.
[23] W. S. Mungall, G. L. Green, G. A. Heavner, R. L. Letsinger, *J. Org. Chem*. **1975**, *40*, 1659.
[24] T. Adachi, Y. Yamad, I. Inoue, M. Saneyoshi, *Synthesis* **1977**, *1*, 45.
[25] J.-K. Chen, R. G. Schultz, D. H. Lloyd, S. M. Gryaznov, *Nucleic Acids Res.* **1995**, *23*, 2661.
[26] N. E. Poopeiko, T. I. Pricota, I. A. Mikhailopulo, *Synlett* **1991**, *4*, 342.

Received April 30, 2001