Experimental

General

The solvents were reagent grade and were distilled and dried by conventional methods before use. The products were purified by flash chromatography on silica gel 60 (Merck 0.063 mm, 230-400 mesh ASTM). NMR spectra were obtained on a Bruker AC 200 and MSL 300 MHz spectrometers. δ-Values are reported in ppm relative to Me_4Si as standard for ^1H NMR (200.13 and 300.13 MHz) and relative to H_3PO_4 as external standard for ^{31}P NMR (80.96 and 121.49 MHz.), as relative to $CFCl_3$ as external standard for ^{19}F NMR (188.15 MHz). The signals are expressed as s (singlet), d (doublet), t (triplet) or m (multiplet). Coupling constants (*J*) are in Hz

General Syntheses

The solution of the appropriate amino alcohol (10 mmol) and DBU (10 mmol) in dry CH_3CN was added at room temperature under N_2 atmosphere to a solution of corresponding *O*-arylophosphite (10 mmol) in dry CH_3CN. The progress of the reaction was monitored by ^{31}P NMR and TLC. When the reaction was complete the reaction mixture was evaporated *in vacuo.* The residue was purified by flash chromatography or destilation.

Synthesis bis(*O*-4-nitrophenyl)-*N*,*N*-diisopropylphosphoramidite 1*:* (Route a) The solution of *N,N*-diisopropyldichlorophosphoramidite (10 mmol) in dry THF (10 ml) was added dropwise at room temperature under a nitrogen atmosphere to a solution of sodium 4-nitrophenolate (20 mmol) in dry THF (50 ml) with stirring for 2h. The sodium chloride was removed by filtration. The filtrate evaporated to dryness and the 3 purified by column chromatography (Et_2O/n-pentane/triethylamine 50:30:5 v\v, Rf: 0.75) Yield 95%. (Route b) The solution of trimethyl(p-nitrophenoxy)silane (20 mmol) in dry THF (20 ml) was added to a solution of *N,N*-diisopropyldichlorophosphoramidite (10 mmol) in dry THF (20 ml) at room temperature. The mixture was stirred for 1h, then trimethylchlorosilane and solvent were removed under reduced pressure to give pure phosphoramidite **1**. Yield 97%.

δ_P (80.96 MHz, $CDCl_3$) 144.8; δ_H (200.13 MHz, $CDCl_3$) 1.01 (12H, d, *J* 6.8 N[CH(CH_3)$_2$]$_2$), 3.46-3.65 (2H, m, N[C*H*(CH_3)$_2$]$_2$), 6.75 (4H, d, *J* 9.13, Ph-H_{ortho}) 7.86 (4H, d, *J* 9.12, Ph-H_{meta}), m.p. 120^0C-122^0C; pale yellow crystals, FAB(M+1) calculated for $C_{18}H_{22}N_3O_6P$: 407.13, found: 408.50.

Synthesis of *O*-[5'-*O*-(*tert*-butyldimethylsilyl)deoxyadenosidin-3'-yl] *O*-(4-nitrophenyl)-*N*,*N*-diisopropylphosphoramidite 3: The solution of the 5'-*O*-(*tert*-butyldimethylsilyl)adenosidine (1.0 mmol) and DBU (1.0 mmol) in dry acetonitrile (15 mL) was added dropwise at room temperature under a nitrogen atmosphere to a solution of bis(*O*-4-nitrophenyl)-*N,N*-diisopropylphosphoramidite **3** (1.1 mmol) in dry acetonitrile (15 mL) with stirring for 16 hrs. The mixture was evaporated to dryness and the resulting residue was purified by column chromatography using CH_2Cl_2:$CH_3C(O)CH_3$ (10:5 v/v) as an eluent to give pure **3**.

Yield 95%; δ_P (121.49 MHz, $CDCl_3$) 147.6, 146.9; δ_H (300.13 MHz, $CDCl_3$) 0.79 (6H, s), 1.11 (9H, s) 1.22, 1.26 (12H, 2d, *J* 6.9, 6.3 C*H*$_3$ of isopropyl), 2.83 (1H, m, H-2'), 3.09 (1H, m, H-2''), 3.43 and 3.61 (4H, m, H-5', 5'' and CH of isopropyl), 4.19-4.31 (1H, m, H-4'), 5.22-5.31 (1H, m, H-3'), 5.95 (2H, br s, NH_2) 6.43 6.87 (1H, dd, *J* 5.2, *J* 8.9, H-1'), 7.36 (2H, d *J* 6.00, 4-NO_2Ph-H_{ortho}), 8.09 (1H, s, H-2), 8. 29 (2H, d, *J* 6.00, 4-NO_2Ph-H_{meta}), 8.78, (1H, s, H-8); δ_C (75.47 MHz, $CDCl_3$) -5.12 (Si-CH_3), 18.38 (Si-C), 23.09, 23.18(J_{PNCC} 7.0, 5.9, *C*H_3 of isoprpyl), 26.00 (Si-C-*C*H_3), 40.12 (C-2'), 45.31, 45.43 (J_{PNC} 6.1, 4.9, *C*H of isopropyl), 64.65 (C-5'), 75.87 (C-3'), 85.65 (C-1'), 86.81 (C-4'), 119.22 (d, *J* 10.52, C-2 of 4-NO_2Ph), 124.01 (C-5), 125.97 (C-3 of 4-NO_2Ph), 136.00 (C-4 of 4-NO_2Ph), 140.82 (C-8), 145.98, (C-4), 150.07 (C-2), 152.32 (C-6), 161.32 (d, *J* 6.85, C-1 of 4-NO_2Ph). FAB(M+1) calculated for $C_{28}H_{44}N_7O_6PSi$: 633.77 found: 634.90.

Synthesis of *O*-[5'-*O*-(*tert*-butyldimethylsilyl)deoxyadenosidin-3'-yl] *N*,*N*-diisopropylfluorophosphoramidite 4: To a solution of aryl phosphoramidite **3** (1.0 mmol) in dry THF (10 mL) was added TBAF (1.2 mmol) at room temperature. After 10 min. tetra-n-butylammonium 4-nitrophenolate was removed by filtration. The filtrate was concentrated *in vacuo* and residue was purified by column chromatography using CH_2Cl_2:CH_3COCH_3 as an eluent to give pure fluorophosphoramidite **4**.

Yield 92%; δ_P (121.49 MHz, $CDCl_3$) 156.0 (d, J_{PF} 1114.9) 158.9(d, J_{PF} 1114.5); δ_F(188.15 MHz, $CDCl_3$) -76.4 (d, J_{PF} 1116.9) -77.0 (d, J_{PF} 1115.6); δ_H (300.13 MHz, $CDCl_3$) 0.81 (6H, s), 1.01 (9H, s), 1.23, 1.25 (12H, 2d, *J*

6.9, 6.3 CH_3 of isopropyl), 2.80 (1H, m, H-2'), 3.19 (1H, m, H-2''), 3.49 and 3.60 (4H, m, H-5', 5'' and CH of isopropyl), 4.22-4.31 (1H, m, H-4'), 5.20-5.33 (1H, m, H-3'), 6.02 (2H, br, NH_2) 6.55 6.88(1H, dd, *J* 5.5, *J* 8.7, H-1'), 8.11 (1H, s, H-2), 8.66 (1H, s, H-8); δ_C (75.47 MHz, $CDCl_3$) -4.88 (Si-CH_3), 17.43 (Si-C), 22.90, 22.88(J_{PNCC} 7.3, 4.9, *C*H_3 of isoprpyl), 26.01 (Si-C-*C*H_3), 40.23 (C-2'), 45.31, 45.43 (J_{PNC} 6.1, 4.9, *C*H of isopropyl), 63.45 (C-5'), 74.59 (C-3'), 84.60, 84.71 (C-1'), 85.40, 85.71 (C-4'), 123.41 (C-5), 139.32 (C-8)145.78 (C-4), 148.68 (C-2), 150.67 (C-6).

FAB(M+1) calculated for $C_{22}H_{40}FN_6O_3PSi$: 514.66, found: 515.89.

***O*-(9 [(2-Hydroxyethoxymethyl)guanin-4'-yl] *O*-(4-nitrophenyl) *N*,*N*-diisopropylphosphoramidite 8:** The solution of the 9-(2-hydroxyethoxymethyl)guanine (1.0 mmol) and DBU (1.0 mmol) in dry acetonitrile (15 mL) was added dropwise at room temperature under a nitrogen atmosphere to a solution of *bis*(*O*-4-nitrophenyl)-*N,N*-diisopropyl-phosphoramidite 3 (1.1 mmol) in dry acetonitrile (15 mL) with stirring for 10 min. The mixture was evaporated to dryness and the resulting residue was purified by column chromatography using CH_2Cl_2:$CH_3C(O)CH_3$ (10:3 v/v) as an eluent to give pure **8** Yield 85 %; δ_P (121.49 MHz, $CDCl_3$) 145.0, 145.5 (1:1); δ_H (300.13 MHz, $CDCl_3$) 1.18 1.21 (12 H, 2d, *J* 7.3, 6.9, CH_3 of isopropyl), 3.37-3.50 (2H, m, CH of isopropyl), 3.69 (2H, m, H-3'), 4.10 (2H, m, H-4'), 5.50 (2H, s, H-1'), 7.14 (2H, d, *J* 6.18, 4-NO_2-Ph-H_{ortho}), 7.90 (1H, s, H-8) 8.40 (2H, d, *J* 6.1,1 4-NO_2-Ph-H_{meta}), δ_C(57.47 MHz, $CDCl_3$) 24.87, 25.54 (J_{PNCC} 8.8, 6.9, CH_3 of isopropyl), 45.81, 46.08 (J_{PNC} 7.5, 6.1, CH of isopropyl), 65.1 (d, J_{COP}=5.9 C-4'), 71.2 (d, J_{CCOP}=7.4, C-3'), 73.5 (C-1'), 112.3 (C-5), 118.44 (d, *J* 10.00, C-2 of 4-NO_2Ph), 121.19 (C-3 of 4-NO_2Ph) 137.7 (C-8), 151.3 (C-4), 158.2 (C-2), 160.31 (d, *J* 6.91, C-1 of 4-NO_2Ph),163.1 (C-6). FAB(M+1) calculated for $C_{20}H_{28}N_7O_6P$: 493.46, found: 494.39

Synthesis of *O*-(5'-*O*-(*tert*-butyldimethylsilyl)deoxyadenosidin-3'-yl) *O*-(3'-*O*-acetylthymidin-5'-yl)-*N*,*N*-diisopropylphosphor-amidite 10. To the solution of *O*-[5'-*O*-(*tert*-butyldimethylsilyl)deoxyadenosidin-3'-yl] *O*-(4-nitrophenyl)-*N,N*-diisopropylphosphoramidite: in dry THF (10 mL) was added DBU (12 mmol) at room temperature. After 10 min the ^{31}P NMR of the reaction mixture exhibited new signal (δ_P: 125.6 ppm and 125.86 ppm) and 3'-*O*-acetylthymidine was added. After 10 hrs. 1,8-diazabicyclo[5,4,0]undec-7-ene 4-

nitrophenolate was removed by filtration. The filtrate was concentrated *in vacuo* and residue was purified by column chromatography using CH_2Cl_2:CH_3COCH_3 (10/3 v/v) as an eluent to give pure *O*-(5'-*O*-(*tert*-butyldimethylsilyl)adenosidin-3'-yl) *O*-(3'-*O*-acetylthymidin-5'-yl)-*N,N*-diisopropylphosphor-amidite 10. Yield 95%; δ_P (121.49 MHz, $CDCl_3$) 149.2, 148.8; δ_H (300.13 MHz, $CDCl_3$) 0.63 (6H, s), 0.91 (9H, s), 1.25, 1.26 (12H, 2d, *J* 6.9, 6.3 CH_3 of isopropyl), 1.73 (3H, s, 5-CH_3), 2.11(3H, s, of Ac) 2.49 (1H, m, H-2' Th), 2.63 (1H, m, H-2" Th), 2.80 (1H, m, H-2' Ad), 3.11 (1H, m, H-2" Ad), 3.50-3.77 (6H, m, H-5', 5" of Th, Ad and CH of isopropyl), 4.19-4.31 (2H, m, H-4', Thy and Ade), 5.22-5.31 (2H, m, H-3',Thy and Ade), 6.01 (2H, br s, NH_2), 6.45-6.47(2 H, m, H-1' Thy and Ade), 7.77 (1H, s) 8.09 (1H, s, H-2), 8.78 (1 H, s, H-8); δ_C (75.47 MHz, $CDCl_3$) -4.73 (Si-CH_3), 14.88 (CH_3 Th), 21.43 [*C*H_3C(O)] 18.31 (Si-C), 22.53, 22.98(J_{PNCC} 7.3, 4.9, CH_3 of isoprpyl), 25.83 (Si-C-*C*H_3), 40.01, 40.23 (C-2', Thy and Ade), 45.31, 45.43 (J_{PNC} 6.1, 4.9, CH of isopropyl), 62.41, 63.77(C-5', Thy and Ade), 74.59, 74.91 (C-3', Thy and Ade), 84.71, 84.73 (C-1', Thy and Ade), 85.11, 85.32 (C-4', Thy and Ade), 111.22 (C,-5 Thy) 123.21 (C-5, Ade), 138.11 (C-8, Ade) 145.99 (C-4, Ade), 148.39 (C-4, Thy), 152.39, 152.55 (C-2, Thy and Ade), 155.67 (C-6, Ade), 164.12 (C-6, Thy), 171.87 (C(O), Ac) FAB(M+1) calculated for $C_{32}H_{53}N_8O_8PSi$: 736.89; found: 738.01.

Synthesis of *O*-(5-*O*-(*tert*-butyldimethylsilyl)deoxyadenosin-3-yl) *O*-(3-*O*-acetylthymidin-5-yl) *O*-(4-nitrophenyl)phosphite 13 To the solution of *O*-[5'-*O*-(*tert*-butyldimethylsilyl)deoxyadenosidin-3'-yl] *O*-(4-nitrophenyl)-*N,N*-diisopropylphosphoramidite in dry THF (10 mL) was added 2,4-dinitrophenol (12 mmol) at room temperature. After 10 min 3'-*O*-acetylthymidine was added. The mixture was stirred 2 h, filtered and the filtrate concentrated to dryness under reduced pressure. The residue was purified by column chromatography using CH_2Cl_2:CH_3COCH_3 as an eluent to give pure phosphite **13**.

Yield 82%; δ_P (121.49 MHz, $CDCl_3$) 135.2, 136.2; δ_H (300.13 MHz, $CDCl_3$), 0.9 (6H, s), 1,00 (9H, s), 2,13 (3H, s, CH_3 of Ac) 2.83-3.00 (2H, m, H-2', Thy and Ade), 3.19-3.33 (2H, m, H-2", Thy and Ade), 3.43-3.61 (4H, m, H-5', 5", Thy and Ad), 4.19-4.31 (2H, m, H-4' of Th and Ad), 5.20-5.36 (2H, m, H-3', Thy and Ade), 5.88 (2H, br s, NH_2) 6.45-6.47(2H, m, H-1', Thy and Ade), 7.13 (2H, d, *J* 9.14, 4-NO_2Ph-H_{ortho}), 8.15 (1 H, s, H-2), 8. 20 (2 H, d, *J* 9.17, 4-NO_2Ph-H_{meta}), 8.80 (1H, s, H-8); δ_C (75.47 MHz, $CDCl_3$) -5.12 (Si-CH_3), 11.84 (CH_3, Thy) 18.38 (Si-C), 26.00 (Si-C-*C*H_3) 34.01, 35.23 (C-2', Thy and Ade), 63.13, 63.45 (C-5', Thy and

Ade), 75.79, 76.99 (C-3', Thy and Ade), 84.66, 84.99 (C-1', Thy and Ade), 85.40, 85.71 (C-4', Thy and Ade), 111.11 (C-5, Thy) 119.20 (d, *J* 10.52, C-2 of 4-NO_2Ph), 123.41 (C-5, Ade), 125.27 (C-3 of 4-NO_2Ph), 136.00 (C-4 of 4-NO_2Ph), 139.99(C-8 Ade), 147.46 (C-4, Ade), 149.67 (C-2, Ade), 150.56 (C-2 Thy) 153.82 (C-6 Ade), 160.52 (d, *J* 6.85, C-1 of 4-NO_2Ph) 164.22 (C-4 Thy) FAB(M+1) calculated for $C_{32}H_{43}N_8O_{11}PSi$ 774.81. found: 775.92

Synthesis of 2-(2,4-dinitrophenoxy)-5,5-dimethyl-1,3,2-dioxaphosphorinane 14: The solution of 2-chloro-5,5-dimethyl-1,3,2-dioxaphosphorinane (10 mmol) in dry THF (10 ml) was added dropwise at room temperature under a nitrogen atmosphere to a solution of 2,4-dinitrophenol (10 mmol) and triethylamine (10 mmol) in dry THF (10 ml). The mixture was stirred 2 h, filtered and the filtrate concentrated to dryness under reduced pressure to afford the 2-(2,4-dinitrophenoxy)-5,5-dimethyl-1,3,2-dioxaphosphorinane **14** (pale yellow crystals) which was directly used without further purification. ^{31}P NMR. δ_P (121.49 MHz, $CDCl_3$): 116.3, δ_H (200.13 MHz, $CDCl_3$) 0.31 (3H, s), 1.02 (3H, s), 3.14-3.20 (2H, m), 4.12-4.18(2H, m), 6.72(1H, d, J=9.13), 7.57 (1H, d, J=9.13), 8.22 (1H, d, J=2.81) MS: m/z(CI): 317.30 (M+1), $C_{11}H_{13}N_2O_7P$ 316.20.

General procedure for preparation of 3-[(5,5-dimethyl-1,3,2-dioxaphosphorinan-2-yl)oxy]-propylamine 15 or 2-[(5,5-dimethyl-1,3,2-dioxaphosphorinan-2-yl)oxy]-propylamine 16: To the solution of 2-(2,4-dinitrophenoxy)-5,5-dimethyl-1,3,2-dioxaphosphorinane (20 mmol) in dry CH_3CN (20 mL) was added a solution of the appropriate aminoalcohol [3-amino-1-propanol or (±)1-amino-2-propanol] (20 mmol) in dry CH_3CN (20 ml). After stirring for 2 h at room temperature, the solvent was removed in vacuo and the residue dissolved in CH_2Cl_2. This solution was washed aqueous solution of sodium bicarbonate, and water. The organic layer was dried over Na_2SO_4, filtered, and concentrated *in vacuo* to give pure 3-[(5,5-dimethyl-1,3,2-dioxaphosphorinan-2-yl)oxy]-propylamine **15** or 2-[(5,5-dimethyl-1,3,2-dioxaphosphorinan-2-yl)oxy]-1-propylamine **16**.

3-[(5, 5-dimethyl-1,3,2-dioxaphosphorinan-2-yl)oxy]-propylamine 15 δ_P (121.49 MHz, $CDCl_3$): 122.0, δ_H (200.13 MHz, $CDCl_3$) 0.69 (3H, s, CH_3), 1.14 (3H, s, CH_3), 2.01 (2H, m), 3.11 (2H, m), 3.67 (4H, m), 3.80 (2H, m), MS: m/z(CI): 208.34(M+1) $C_8H_{18}NO_3P$ 207.21

2-[(5, 5-dimethyl-1,3,2-dioxaphosphorinan-2-yl)oxy]-propylamine 16 δ_P (121.49 MHz, $CDCl_3$): 122.6, δ_H (200.13 MHz, $CDCl_3$) 0.59 (3H, s), 0.94 (3H, s), 1.19 (3H, d, *J* 6.04), 1.60 (2H, br s, NH_2) 2.81 (2H, m), 3.57(2H, m), 3.89 (3H, m) MS: m/z(CI):208.09 (M+1) $C_8H_{18}NO_3P$ 207.21.

^{31}P NMR study of the reaction of bis (*O*-4-nitrophenyl)-*N*,*N*-diisopropylphosphoramidite 1 with 3',5'-*O*,*O*-di(*tert*-butyldimethylsilyl) 2'-deoxyadenosine 5. A rection was carried out in an NMR tube (5mm x 180 mm) at room temperature. The 3',5'-*O*,*O*-di(*tert*-butyldimethylsilyl) 2'-deoxyadenosine (9.58 mg 20 μmol) was dried by repeated coevaporation with dry pyridine and dissolved in CD_3CN (400 μl) and DBU (3.04 mg 20 μmol) was added. The mixture was transferred into an NMR sample tube. After 10 min bis (*O*-4-nitrophenyl)-*N*,*N*-diisopropylphosphoramidite (8.16 mg 20 μmol) was added. Progress of the reaction was followed by ^{31}P NMR spectroscopy (for the results, see in the text).

^{31}P NMR study of the reaction of *O*-[5'-*O*-(*tert*-butyldimethylsilyl)deoxyadenosidin-3'-yl] *O*-(4-nitrophenyl)-*N*,*N*-diisopropylphosphoramidite 3 with DBU. A reaction was carried out in an NMR tube (5mm x 180 mm) at room temperature. The *O*-[5'-*O*-(*tert*-butyldimethylsilyl)deoxyadenosidin-3'-yl] *O*-(4-nitrophenyl)-*N*,*N*-diisopropylphosphoramidite (12.6 mg 20 μmol) was dried by repeated coevaporation with dry pyridine and dissolved in CD_3CN (400 μl). The mixture was transferred into an NMR sample tube, and DBU (2.5 mg, 2.46 μl, 20 μmol) was added. Monitoring of the reaction mixture by ^{31}P NMR indicated after 10 min, the signal of **3** (147.4, 146.5) disappeared and the new signals were observed in the high-field region at 127.39 ppm and 128.11 ppm.To the mixture was added 3'-*O*-acetylthymidine (dried by repeated coevaporation with dry pyridine). After 10 min, the signals of **11** was readily converted to signals of *O*-(5'-*O*-(*tert*-butyldimethylsilyl)deoxyadenosidin-3'-yl) *O*-(3'-*O*-acetylthymidin-5'-yl)-*N*,*N*-diisopropylphosphoramidite **10** (149.0 ppm, 148.5 ppm).