PYRIMIDINE ACYCLIC NUCLEOTIDE ANALOGUES WITH AROMATIC SUBSTITUENTS IN C-5 POSITION

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NH_2-protected 5-phenylcytosine and its derivatives **2a**–**2d** were treated with (2*S*)-2-[(trityloxy)methyl]oxirane (**3**) followed by etherification with diisopropyl [(tosyloxy)methyl]phosphonate (**5**) in the presence of sodium hydride. The intermediary phosphonate esters **6** were debenzoylated and subsequently transformed to free phosphonic acids, i.e. (*S*)-1-[3-hydroxy-2-(phosphonomethoxy)propyl]-5-phenylcytosine (5-phenyl-HPMPC) derivatives (**8a**–**8d**) by the action of bromotrimethylsilane and subsequent hydrolysis. Deamination of these compounds with 3-methylbutyl nitrite afforded corresponding (*S*)-1-[3-hydroxy-2-(phosphonomethoxy)propyl]-5-phenyluracil (5-phenyl-HPMPU) derivatives (**9a**–**9d**). *R*-Enantiomers **14** and **15** were prepared analogously starting from (2*R*)-2-[(trityloxy)methyl]oxirane. 5-Benzyl-, 5-[(1-naphthyl)methyl]- and 5-[(2-naphthyl)methyl]HPMPU (**24a**–**24c**) and -HPMPC (**25a**–**25c**) were synthesized from appropriate 5-arylmethyl-4-methoxypyrimidin-2(1*H*)-ones similarly as described for 5-phenyl derivatives. Antiviral activity was found for (*S*)-1-[3-hydroxy-2-(phosphonomethoxy)propyl]-5-phenyluracil (**9a**) (HSV-1 and HSV-2) and (*R*)-1-[3-hydroxy-2-(phosphonomethoxy)propyl]-5-phenylcytosine (**14**) (cytomegalovirus and varicella-zoster virus), both tested in cell cultures. Some of the 5-phenyluracil derivatives possessed inhibitory activity against thymidine phosphorylase from SD-lymphoma.

Keywords: Acyclic nucleotide analogues; Acyclic nucleoside phosphonates; Pyrimidines; Antivirals; Thymidine phosphorylase inhibitors; Cidofovir; Cytosine.

Acyclic nucleotide analogues (ANPs) are compounds of considerable importance due to their broad spectrum of antiviral activities[1]. An important group of these compounds are (*S*)-[3-hydroxy-2-(phosphonomethoxy)propyl] (HPMP) derivatives of purine and pyrimidine bases: adenine and diaminopurine (HPMPA, HPMPDAP) and, in particular, the cytosine derivative (HPMPC, cidofovir[2]), an active constituent of Vistide™. This drug was approved for the treatment of cytomegalovirus retinitis in AIDS patients[3]. At the present time, it is important particularly for the treatment of severe cases of (malignizing) papillomatoses (anogenital, laryngeal)[4], progressive

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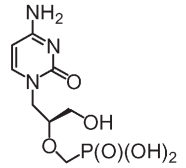
multifocal leukoencephalopathy (caused by JC virus)[5] adenovirus infections[6] and some rather obscure severe infections caused by poxviruses (vaccinia[7], orf, molluscum contagiosum). The attractivity of cidofovir is dramatically enhanced by its supreme activity against smallpox virus[8] and related monkeypox virus[9]; both of these highly infectious viruses can be easily cultivated and purposely used in a bioterrorist attack.



(*S*)-HPMPC (cidofovir)

These facts support further investigation of variously modified cidofovir derivatives. An intensive attention is paid to the development of neutral ester prodrugs[10] and, at present, we focus our attention also to preparation of analogues modified in the cytosine moiety (analogues substituted in positions C-5 and N^4, refs[11,12]).

In this paper, we describe preparation of HPMPC substituted in position 5 with various aromatic systems as compounds for antiviral screening. Simultaneously, we prepared also their uracil counterparts (HPMPU derivatives). ANPs derived from uracil and thymine are usually antivirally inactive[1]; their investigation is directed to study of other biological properties, in particular inhibitory effects on thymidine phosphorylase[13], an enzyme catalyzing phosphorolysis of thymidine to thymine and 2-deoxy-D-ribose 1-phosphate.

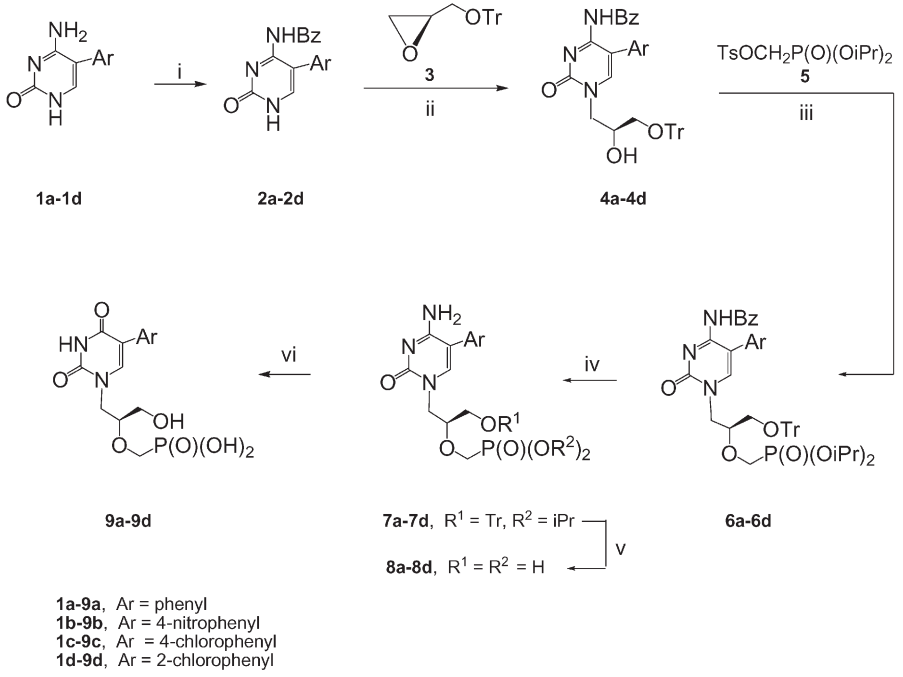
As to nucleobases themselves, some attention in the recent years, was paid to substituted 5-phenylcytosines and intermediates formed in their preparation as antiviral agents: 2-[(carboxymethyl)sulfanyl-5-(4-chlorophenyl)pyrimidin]-4-amine is active in vitro against vaccinia virus[14], some activity was also found for its *o*- and *m*-isomer. 5-Phenylcytosine and its derivatives (*p*-amino, *p*-chloro and *m*-chloro) were studied as inhibitors of WEE (Western equine encephalomyelitis) virus. In in vivo experiments all these compound were inactive, probably due to their insolubility in water[14]. Nucleosides prepared from 5-phenylcytosine and 5-phenyluracil are also in most cases antivirally inactive[15,16]. 5-Benzyluracil nucleosides and their acyclic analogues were found to be inhibitors of uridine phosphorylase, an enzyme responsible for undesirable phosphorolysis of pyrimidine

nucleoside analogues used in cancer chemotherapy[17,18]. Pyrimidine bases and their nucleoside and/or nucleotide analogues substituted in position 5 with bulky naphthylmethyl group have not been described yet.

RESULTS AND DISCUSSION

Our approach to 5-aryl HPMPC and HPMPU analogues is based on utilization of appropriately substituted 5-phenylcytosines (**1**). These bases are accessible from corresponding phenylacetonitriles by the process involving formylation, transformation of formyl derivatives to enol ethers and their cyclization with thiourea to 5-phenyl-2-thiocytosines[19,20]. Their reaction with chloroacetic acid followed by hydrolysis with hydrochloric acid affords corresponding 5-phenylcytosines. 5-(4-Nitrophenyl)cytosine (**1b**) was prepared by nitration of 5-phenylcytosine (**1a**)[20].

The amino groups in 5-phenylcytosines **1** were protected by benzoylation with benzoic anhydride in DMF (Scheme 1). This step is useful due to a very

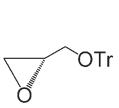


(i) Benzoic anhydride, DMF, r.t.; (ii) DMF, Cs_2CO_3, 100 °C; (iii) NaH, DMF, r.t.; (iv) NH_3/CH_3OH, 4 °C; (v) $(CH_3)_3SiBr$, CH_3CN, r.t; (vi) 3-methylbutyl nitrite, 80% acetic acid, r.t.

SCHEME 1

pure solubility of free phenylcytosines. The following step, base-catalyzed reaction with (2*S*)-2-[(trityloxy)methyl]oxirane (**3**) thus proceeds with high yields in contrast to reactions performed with unprotected cytosine components which are not quantitative. The nucleophilic opening of an oxirane ring proceeded regiospecifically and afforded in all cases N^1-substituted intermediates **4** which gave, on treatment with diisopropyl [(tosyloxy)methyl]-phosphonate (**5**) in the presence of excess sodium hydride in dimethylformamide, fully protected phosphonates **6**. Their partial deprotection to phosphonates **7** with free amino group, followed by simultaneous removal of trityl and diisopropyl ester groups with bromotrimethylsilane and subsequent hydrolysis afforded the free phosphonic acids: 5-phenyl-HPMPC (**8a**) and its substituted derivatives (**8b**–**8d**) as final products. These compounds can be also transformed to their uracil counterparts: (*S*)-1-[3-hydroxy-2-(phosphonomethoxy)propyl]-5-phenyluracil (5-phenyl-HPMPU, **9a**) and its 4-nitro, 4-chloro and 2-chloro derivatives (**9b**–**9d**); deamination reaction was performed with 3-methylbutyl nitrite (isoamyl nitrite) in acetic acid.

The anti-herpes virus activity found for (*S*)-1-[3-hydroxy-2-(phosphonomethoxy)propyl]-5-phenyluracil (**9a**) substantiates our effort to prepare for comparison also the corresponding (*R*)-enantiomer (**15**) and 5-phenyluracil nucleotide analogues with modified aliphatic moiety, in particular 5-phenyl-1-[(*R*)-2-(phosphonomethoxy)propyl]uracil, a compound belonging to the PMP-series[1]. The (*R*)-enantiomer **15** was prepared from (2*R*)-2-[(trityloxy)methyl]oxirane (**10**) and N^4-benzoyl-5-phenylcytosine (**2a**) via intermediates **11**–**14** by the same procedure as described for (*S*)-enantiomer **9a**.



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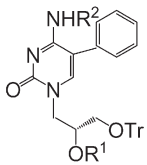
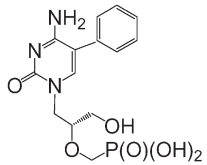


11, R^1 = OH, R^2 = Bz
12, R^1 = $CH_2P(O)(OiPr)_2$, R^2 = Bz
13, R^1 = $CH_2P(O)(OiPr)_2$, R^2 = H



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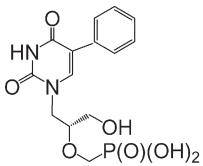

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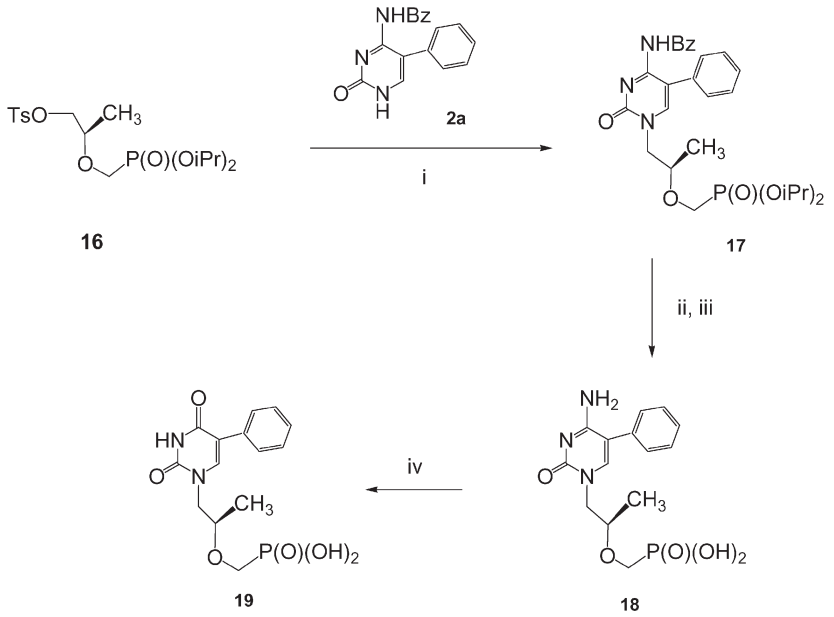
ANPs of the (*R*)-2-(phosphonomethoxy)propyl series are usually prepared by condensation of an appropriate base component with (*R*)-2-[(diisopropoxyphosphoryl)methoxy]propyl tosylate (**16**) under basic conditions[21]. The reaction of unprotected 5-phenylcytosine with **16** in DMF was not successful,

probably due to very low solubility of 5-phenylcytosine. The heating of N^4-benzoyl-5-phenylcytosine with a large excess of **16** in the presence of cesium carbonate gave the desired compound **17** accompanied by several other products, partly deprotected phosphonates including monoisopropyl esters (as determined by paper electrophoresis). This mixture of products afforded, after treatment with methanolic ammonia and deprotection of ester groups with bromotrimethylsilane, 5-phenyl-1-[(*R*)-2-(phosphonomethoxy)-propyl]cytosine (**18**) as a single reaction product. Its deamination with isoamyl nitrite afforded 5-phenyl-1-[(*R*)-2-(phosphonomethoxy)propyl]uracil (**19**) (Scheme 2).

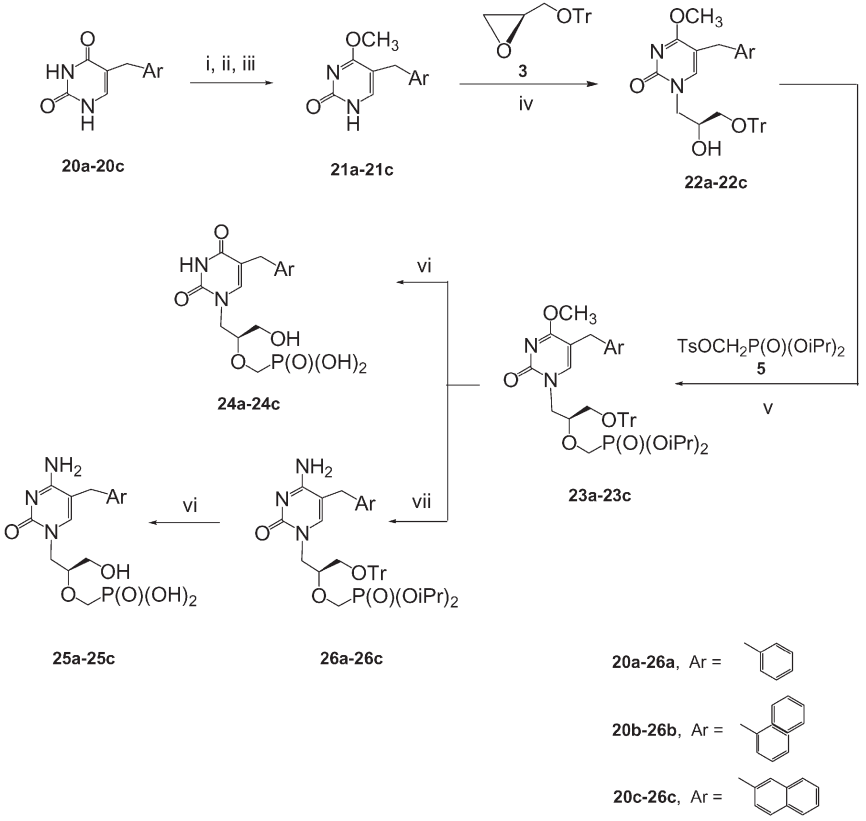


(i) DMF, Cs_2CO_3, 120 °C; (ii) NH_3/CH_3OH, 4 °C; (iii) $(CH_3)_3SiBr$, CH_3CN, r.t.; (iv) 3-methylbutyl nitrite, 80% acetic acid, r.t.

SCHEME 2

For syntheses of 5-benzyl- and both 5-(naphthylmethyl)-HPMPU and -HPMPC derivatives, we selected an approach, which is in principle analogous to preparation of 5-phenyl derivatives: nucleophilic opening of (2*S*)-2-[(trityloxy)methyl]oxirane (**3**) with appropriate base component, followed by the treatment with diisopropyl [(tosyloxy)methyl]phosphonate (**5**) to give fully protected phosphonate esters **23** (Scheme 3). In this case 5-substituted uracils were used as starting bases. The preparation of

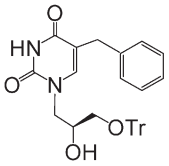
5-benzyluracil (**20a**), 5-(1-naphthylmethyl)uracil (**20b**) and 5-(2-naphthylmethyl)uracil (**20c**) was performed from ethyl 3-phenylpropanoate or ethyl 3-naphthylpropanoates according to refs[22,23]. Their transformation to 5-(arylmethyl)-4-methoxypyrimidin-2(1*H*)-ones **21** consists in the treatment with $POCl_3$ giving 2,4-dichloropyrimidines, reaction with sodium methoxide leading to 2,4-dimethoxypyrimidines and their final transformation to 4-methoxypyrimidin-2(1*H*)-ones **21** by the action of acetyl chloride followed by sodium methoxide. This procedure, developed originally for uracil and thymine protection[24] was found useful for all types of 5-substituted uracils; recently we used this process for preparation of



(i) $POCl_3$, *N*,*N*-dimethylaniline; (ii) CH_3ONa; (iii) CH_3COCl, followed by CH_3ONa; (iv) DMF, Cs_2CO_3, 80 °C; (v) NaH, THF, r.t.; (vi) $(CH_3)_3SiBr$, CH_3CN, r.t.; (vii) NH_3/CH_3OH, 110 °C, autoclave

SCHEME 3

5-ethyl-4-methoxypyrimidin-2(1*H*)-one[11]. Reactions of 5-substituted 4-methoxypyrimidin-2(1*H*)-ones with oxirane **3** were performed in DMF under Cs_2CO_3 catalysis at 80 °C. An increase in temperature (up to 120 °C) led to deprotection of 4-methoxy group (compound **27**), simultaneously with the formation of undesired disubstituted derivative **28**. Phosphonate diesters **23** were deprotected in one step with bromotrimethylsilane in acetonitrile and subsequent hydrolysis to 5-benzyl-1-[(*S*)-3-hydroxy-2-(phosphonomethoxy)propyl]uracil (**24a**) and its 5-(naphthylmethyl) analogues (**24b**, **24c**) as final products for biological testing. Their cytosine counterparts **25a**–**25c** were prepared from 4-methoxypyrimidin-2(1*H*)-one intermediates **23a**–**23c** by high-pressure reaction with ammonia followed by deprotection with bromotrimethylsilane.



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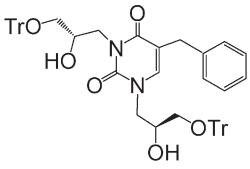

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Biological Activity

All target compounds were tested for antiviral activity in vitro in cell cultures (E_6SM cell culture for herpesviruses, human embryonic lung cells for cytomegalovirus and varicella-zoster virus). A selective anti-herpes virus activity was found for (*S*)-1-[3-hydroxy-2-(phosphonomethoxy)propyl]-5-phenyluracil (**9a**) while the corresponding 5-phenylcytosine derivative **8a** was completely inactive. Interestingly, an antiviral activity was found also for (*R*)-configurated HPMP-5-phenylcytosine **14**, in spite of the fact that (*R*)-HPMP derivatives are usually inactive. Activity data of these compounds are shown in Table I. All other compounds described in this paper were antivirally inactive.

All 5-phenyluracil derivatives were tested for inhibitory activity against thymidine phosphorylase (E.C.2.4.2.4.) from SD-lymphoma. This investigation was performed under conditions described recently in the study of TP inhibition with diverse thymine (phosphonomethoxy)alkyl derivatives: 100 μM [^{3}H]-2′-deoxythymidine, 250 μM P_i, tested compound 10 μmol l^{-1}, pH 6.7; an appropriate amount of enzyme and 10 min incubation at 37 °C (ref.[13]). Inhibitory activity was found at 1-[(*R*)-3-hydroxy-2-(phosphono-

methoxy)propyl]-5-phenyluracil (**15**, V_i/V_0 = 0.25), the activity of other compounds was only marginal: V_i/V_0 = 0.53 at 5-phenyl-1-[(*R*)-2-(phosphonomethoxy)propyl]uracil (**19**) and V_i/V_0 = 0.72 at 1-(*S*)-[3-hydroxy-2-(phosphonomethoxy)propyl]-5-(4-nitrophenyl)uracil (**9b**).

TABLE I
Antiviral activity of compounds **9a** and **14** against cytomegalovirus (CMV), varicella-zoster virus (VZV) and herpes simplex viruses (HSV-1, HSV-2) compared to cidofovir (HPMPC) and ganciclovir

Compound	Antiviral activity[a] EC_{50}, μg/ml						MCC[b] μg/ml
	CMV AD 169 strain	CMV Davis strain	TK^+ VZV OKA strain	TK^- VZV 07/01 strain	HSV-1 (KOS)	HSV-2 (G)	
9a	76	55	>100	>100	9.6	9.6	>400
14	2.2	0.8	4.4	1.5	240	48	>400
Cidofovir	0.13	0.35	0.20	0.05	0.16	0.54	>400
Ganciclovir	0.64	1.7	–	–	0.0064	0.0768	>400

[a] Effective concentration required to reduce virus plaque formation by 50%. [b] Minimum cytotoxic concentration that causes a microscopically detectable alteration of cell morphology.

None of the compounds presented in this study exhibited cytostatic activity in vitro tested in mouse lymphocytic leukemia L1210 cells, CCRF-CEM T lymphoblastoid cells, human promyelocytic leukemia HL-60 cells and human cervix carcinoma HeLa cells.

EXPERIMENTAL

Unless stated otherwise, solvents were evaporated at 40 °C/2 kPa and compounds were dried at 13 Pa. Melting points were determined on a Kofler block and are uncorrected. Analytical TLC were performed on silica gel 60 F_{254} plates (Merck KGaA, Darmstadt, Germany); chromatographic systems are described in text. Column chromatography was performed on silica gel 60 μm (Fluka) or aluminum oxide (50–150 μ, pH 7.0 ± 0.5, Fluka). Reverse phase HPLC separations were performed on a Waters Delta 600 instrument with a Waters 2487 Dual λ Absorbance Detector using columns XTerra® RP_{18} (3.9 × 150 mm, analytical column) and Luna Phenomenex® C-18 (21 × 250 mm, preparative column). ^{1}H NMR spectra were measured on a Varian Unity 500 instrument (at 500 MHz) in DMSO-d_6 solutions (referenced to the solvent signal at δ 2.50) or in D_2O solutions with internal standard sodium 3-(trimethylsilyl)propane-1-sulfonate (DSS). ^{1}H NMR chemical shifts (δ, ppm) and coupling constants (*J*, Hz) were obtained by first-order analysis of the spectra. ^{13}C NMR spectra were recorded on the same instrument (at 125.7 MHz) using APT pulse sequence in DMSO-d_6 (ref-

erenced to the solvent signal δ 39.70). The numbering system for assignment of NMR signals is outlined in Fig. 1. Mass spectra were measured on a ZAB-EQ (VG Analytical) spectrometer using FAB (ionization with xenon, accelerating voltage 8 kV, glycerol matrix) or ESI. Optical rotations were measured on Autopol IV polarimeter (Rudolph Research Analytical, U.S.A.) at 20 °C, $[\alpha]_D$ values are given in 10^{-1} deg cm^2 g^{-1}.

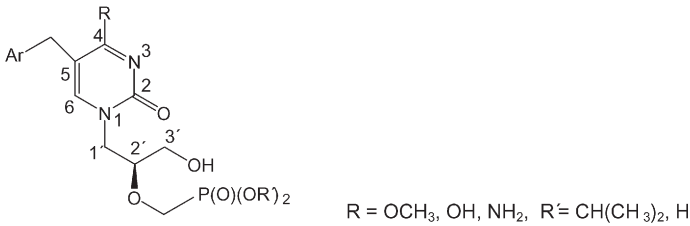

FIG. 1
General numbering scheme for assignment of NMR signals

Materials and Solvents

Most chemicals and ion-exchange resins (Dowex 50WX8-200 and Dowex 1X2-400) were purchased from Sigma–Aldrich (Czech Republic). Dimethylformamide and acetonitrile were dried by distillation from CaH_2 (DMF in vacuo) and stored over molecular sieves (4Å). Diisopropyl [(tosyloxy)methyl]phosphonate was prepared by the described procedure[25]. (2*S*)-2-[(Trityloxy)methyl]oxirane was purchased from DAISO Co. Ltd (Japan).

Preparation of 5-Aryl-N^4-benzoylcytosines **2a**–**2d**. General Procedure

Benzoic anhydride (6.8 g, 30 mmol) and 4-(dimethylamino)pyridine (50 mg) were added to a suspension of an appropriate 5-phenylcytosine **1** (20 mmol) in DMF (250 ml). The mixture was stirred at room temperature for 3 days, then diluted with ethanol (50 ml) and evaporated. The residue was coevaporated with toluene (2 × 50 ml) and subsequently refluxed with ethanol (150 ml) for 15 min. After cooling to room temperature, ether (50 ml) was added, the crystalline product collected by suction, washed with ether and dried in vacuo.

N^4-Benzoyl-5-phenylcytosine (**2a**). Yield 4.1 g (70%), white crystals. M.p. 237 °C. For $C_{17}H_{13}N_3O_2$ (291.3) calculated: 70.09% C, 4.50% H, 14.42% N; found: 69.68% C, 4.38% H, 14.29% N. FAB MS, *m/z* (%): 292 (100) [M + H]. ^{1}H NMR (DMSO-d_6): 13.35 brs, 1 H and 12.03 brs, 1 H (NH); 8.0 d, 2 H (H-arom.); 7.60–7.30 m, 8 H (H-arom.); 7.86 s, 1 H (H-6). ^{13}C NMR (DMSO-d_6): 172.54 (Bz); 159.88 (C-4); 148.07 (C-2); 143.42 (C-6); 137.25 (Bz); 133.50 (Ph); 132.55 (Bz); 129.67, 2 C and 129.44, 2 C (Ph); 128.44, 2 C and 128.02 (Bz); 127.56 (Ph); 113.44 (C-5).

N^4-Benzoyl-5-(4-nitrophenyl)cytosine (**2b**). Yield 5.0 g (74%), yellowish crystals. M.p. 284–288 °C. For $C_{17}H_{12}N_4O_4$ (336.3) calculated: 60.71% C, 3.60% H, 16.66% N; found: 60.37% C, 3.72% H, 16.68% N. FAB MS, *m/z* (%): 337 (4) [M + H], 233 (16) [M – benzoyl + 2 H]. ^{1}H NMR (DMSO-d_6): broad signals due to insufficient solubility of compound.

N^4-Benzoyl-5-(4-chlorophenyl)cytosine (**2c**). Yield 5.8 g (89%), white crystals. M.p. 308 °C. For $C_{17}H_{12}ClN_3O_2$ (325.8) calculated: 62.68% C, 3.71% H, 10.88% Cl, 12.90% N; found:

62.78% C, 3.70% H, 10.64% Cl, 13.00% N. FAB MS, *m/z* (%): 326 (18) [M + H], 105 (28) [benzoyl]. ^{1}H NMR (DMSO-d_6): 13.30 brs, 1 H and 12.10 brs, 1 H (NH); 8.03 brd, 2 H (H-arom.); 7.91 s, 1 H (H-6); 7.66 brd, 2 H, 7.55 t, 1 H, 7.49 d, 2 H and 7.45 t, 2 H (H-arom.). ^{13}C NMR (DMSO-d_6): 178.41 (Bz); 159.71 (C-4); 148.91 (C-6); 143.91 (C-2); 137.05 (Ph); 132.59 (Bz); 132.19, 131.36, 2 C and 129.42, 2 C (Ph); 128.50, 2 C and 128.02, 2 C (Bz); 112.13 (C-5).

N^4*-Benzoyl-5-(2-chlorophenyl)cytosine* (**2d**). The crude product was chromatographed in toluene–ethyl acetate (1:1). Yield 900 mg (14%), white crystals. M.p. 236–238 °C. FAB MS, *m/z* (%): 326 (100) [M + H], 105 (74) [benzoyl]. ^{1}H NMR (DMSO-d_6): 13.04 brs, 1 H and 11.99 brs, 1 H (NH); 7.87 m, 2 H (H-arom.); 7.85 s, 1 H (H-6); 7.60–7.36 m, 7 H (H-arom.).

Reaction of (2*S*)-2-[(Trityloxy)methyl]oxirane (**3**) with 5-Aryl-*N*^{4}-benzoylcytosines **2a**–**2d**. General Procedure

Compound **3** (2.72 g, 8.6 mmol) and cesium carbonate (250 mg, 0.76 mmol) were added to a stirred suspension of benzoylated 5-arylcytosine **2** (8 mmol) in DMF (50 ml). The mixture was heated at 100 °C until the conversion was complete (6–12 h, TLC control). The mixture was cooled to room temperature, evaporated and the residue coevaporated with toluene (2 × 50 ml). The crude product was purified by chromatography on silica gel in system toluene–ethyl acetate (3:1) containing 1% triethylamine.

N^4*-Benzoyl-1-[(S)-2-hydroxy-3-(trityloxy)propyl]-5-phenylcytosine* (**4a**). Yield 4.0 g (82%) of a white foam. $[\alpha]_D$ –28.9 (*c* 0.802, $CHCl_3$). For $C_{39}H_{33}N_3O_4$ (607.7) calculated: 77.08% C, 5.47% H, 6.91% N; found: 77.10% C, 5.53% H, 6.54% N. FAB MS, *m/z* (%): 608 (58) [M + H], 366 (10) [M – trityl + 2 H], 243 (100) [trityl], 105 (38) [benzoyl]. ^{1}H NMR (DMSO-d_6): 13.55 s, 1 H (NH); 8.08 d, 2 H (H-arom.); 8.04 s, 1 H (H-6); 7.59 d, 2 H, 7.56 t, 1 H, 7.46 t, 3 H, 7.39 d, 6 H, 7.31 t, 6 H and 7.24 m, 5 H (H-arom.); 5.44 d, 1 H, *J*(OH,2′) = 5.5 (OH); 4.11 m, 2 H (H-1′a, H-2′); 3.82 dd, 1 H, *J*(1′b,2′) = 8.1, *J*(gem) = 13.2 (H-1′b); 3.08 br dd, 1 H, *J*(3′a,2′) = 4.4, *J*(gem) = 9.7 (H-3′a); 2.94 dd, 1 H, *J*(3′b,2′) = 5.5 (H-3′b). ^{13}C NMR (DMSO-d_6): 178.475 (C=O); 158.98 (C-4); 147.945 (C-6); 147.84 (C-2); 143.82, 3 C (Tr); 137.15 (Bz); 133.11 (Ph); 132.59, 129.47, 2 C and 129.27, 2 C (Bz); 129.06, 2 C (Ph); 128.38, 8 C (Ph, Tr); 128.06, 6 C (Tr); 127.18, 3 C (Tr); 125.47 (Ph); 113.16 (C-5); 86.185 (Tr); 66.99 (C-2′); 66.18 (C-3′); 52.72 (C-1′).

N^4*-Benzoyl-1-[(S)-2-hydroxy-3-(trityloxy)propyl]-5-(4-nitrophenyl)cytosine* (**4b**). Yield 2.9 g (56%), yellowish crystals. M.p. 233 °C. $[\alpha]_D$ –19.3 (*c* 0.427, $CHCl_3$). For $C_{39}H_{32}N_4O_6$ (652.7) calculated: 71.77% C, 4.94% H, 8.58% N; found: 71.72% C, 4.93% H, 8.49% N. FAB MS, *m/z* (%): 653 (17) [M + H], 243 (100) [trityl], 105 (72) [benzoyl]. ^{1}H NMR (DMSO-d_6): 13.50 brs, 1 H (NH); 8.29 m, 2 H (H-arom.); 8.24 s, 1 H (H-arom.); 8.03 m, 2 H, 7.87 m, 2 H, 7.57 t, 1 H, 7.47 t, 2 H, 7.38 d, 6 H, 7.31 t, 6 H and 7.24 t, 3 H (H-arom.); 5.46 d, 1 H, *J*(OH,2′) = 5.7 (OH); 4.17 dd, 1 H, *J*(1′a,2′) = 3.2, *J*(gem) = 12.7 (H-1′a); 4.11 m, 1 H (H-2′); 3.83 dd, 1 H, *J*(1′b,2′) = 8.4 (H-1′b); 3.07 dd, 1 H, *J*(3′a,2′) = 4.5, *J*(gem) = 9.4 (H-3′a); 2.94 dd, 1 H, *J*(3′b,2′) = 5.8 (H-3′b). ^{13}C NMR (DMSO-d_6): 167.50 (Bz); 156.80 br (C-4); 147.60 br (C-6); 147.20 br (C-2); 146.46, 2 C (NO_2-Ph); 143.81, 3 C (Tr); 132.74 (Bz); 129.37, 2 C (NO_2-Ph); 128.59, 4 C (Bz); 128.38, 6 C, 128.09, 6 C and 127.23, 3 C (Tr); 123.40, 2 C (NO_2-Ph); 110.98 (C-5); 86.20 (Tr); 66.87 (C-2′); 66.225 (C-3′); 53.34 br (C-1′).

N^4*-Benzoyl-5-(4-chlorophenyl)-1-[(S)-2-hydroxy-3-(trityloxy)propyl]cytosine* (**4c**). Yield 3.8 g (74%), white foam. $[\alpha]_D$ –26.3 (*c* 0.426, $CHCl_3$). For $C_{39}H_{32}ClN_3O_4$ (642.2) calculated: 72.95% C, 5.02% H, 5.52% Cl, 6.54% N; found: 73.06% C, 5.29% H, 5.49% Cl, 6.38% N.

FAB MS, *m/z* (%): 642 (1) [M^+], 400 (30) [M – trityl + H], 243 (20) [trityl], 105 (100) [benzoyl]. ^{1}H NMR (DMSO-d_6): 13.50 br s, 1 H (NH); 8.07 s, 1 H (H-6); 8.03 m, 2 H, 7.57 t, 1 H, 7.50 m, 2 H, 7.47 t, 2 H, 7.37 d, 6 H, 7.30 t, 6 H and 7.23 m, 5 H (H-arom.); 5.44 d, 1 H, *J*(OH,2′) = 5.6 (OH); 4.14 dd, 1 H, *J*(1′a,2′) = 4.0, *J*(gem) = 13.2 (H-1′a); 4.09 m, 1 H (H-2′); 3.81 dd, 1 H, *J*(1′b,2′) = 8.1 (H-1′b); 3.06 dd, 1 H, *J*(3′a,2′) = 4.6, *J*(gem) = 9.4 (H-3′a); 2.93 dd, 1 H, *J*(3′b, 2′) = 5.7 (H-3′b). ^{13}C NMR (DMSO-d_6): 167.0 (Bz); 158.0 br (C-4); 148.0 br (C-6); 147.96 (C-2); 144.30 (Cl-Ph); 143.84, 3 C (Tr); 132.68 (Bz); 132.29 br and 129.40, 4 C (Cl-Ph); 128.57–126.83 (arom. C); 111.89 (C-5); 86.20 (Tr); 66.93 (C-2′); 66.24 (C-3′); 53.0 br (C-1′).

N^4-Benzoyl-5-(2-chlorophenyl)-1-[(S)-2-hydroxy-3-(trityloxy)propyl]cytosine (**4d**). Yield 4.1 g (80%), white foam. $[\alpha]_D$ –30.3 (*c* 0.321, $CHCl_3$). For $C_{39}H_{32}ClN_3O_4$ (642.2) calculated: 72.95% C, 5.02% H, 5.52% Cl, 6.54% N; found: 72.43% C, 4.89% H, 5.76% Cl, 6.31% N. ^{1}H NMR (DMSO-d_6): 13.50 br s, 1 H (NH); 8.00 br d, 2 H (H-arom., H-6); 7.50–7.10 m, 23 H (H-arom.); 4.16 dd, 1 H, *J*(1′a,2′) = 2.0, *J*(gem) = 13.8 (H-1′a); 4.14 m, 1 H (H-2′); 3.73 dd, 1 H, *J*(1′b,2′) = 7.2 (H-2′b); 3.30 dd, 1 H, *J*(3′a,2′) = 4.6, *J*(gem) = 9.8 (H-3′a); 3.20 dd, 1 H, *J*(3′b,2′) = 6.0 (H-3′b), 2.80 br, 1 H (OH). ^{13}C NMR (DMSO-d_6): 167.0 br (Bz); 158.79 (C-2); 150.79 (C-2); 146.27 (C-2); 143.33, 3 C ((Tr); 134.50 and 132.63 (Cl-Ph); 132.36 (Bz); 131.50 and 129.89 (Cl-Ph); 129.66, 2 C and 129.60, 2 C (Bz); 128.47, 6 C, 128.03, 6 C and 127.32, 3 C (Tr); 126.46 (Cl-Ph); 112.0 br (C-5); 87.16 (Tr); 69.25 (C-2′); 64.78 (C-3′); 52.45 (C-1′).

Reaction of Compounds **4** with Diisopropyl [(Tosyloxy)methyl]phosphonate (**5**)

A solution of phosphonate **5** (2.0 g, 5.7 mmol) and appropriate trityl derivative **4a**–**4d** (3.9 mmol) in DMF (100 ml) was cooled to –20 °C and 60% sodium hydride dispersion (470 mg, 11.8 mmol) was added under stirring. The mixture was warmed slowly to room temperature, stirred for additional 20 h and then neutralized with acetic acid to pH 7.0 and evaporated. The residue was coevaporated with toluene (100 ml) and chromatographed on silica gel (400 ml) in the system described below.

N^4-Benzoyl-1-{(S)-2-[(diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}-5-phenylcytosine (**6a**). Chromatographed in system ethyl acetate–toluene (2:1). Yield 2.4 g (78%), white foam. $[\alpha]_D$ –53.8 (*c* 0.614, $CHCl_3$). For $C_{46}H_{48}N_3O_7P$ (785.9) calculated: 70.30% C, 6.16% H, 5.35% N, 3.94% P; found: 70.10% C, 6.20% H, 5.18% N, 4.00% P. FAB MS, *m/z* (%): 786.5 (7) [M + H], 544 (10) [M – trityl + 2 H], 243 (100) [trityl]. ^{1}H NMR (DMSO-d_6): 13.55 s, 1 H (NH); 8.07 d, 2 H (H-arom.); 8.03 s, 1 H (H-6); 7.64 d, 2 H, 7.55 t, 1 H and 7.45–7.24 m, 20 H (H-arom.); 4.52 m, 2 H (P-OCH); 4.10–4.00 m, 3 H (H-1′, H-2′); 3.83 dd, 1 H, *J*(P,CH_a) = 9.0, *J*(gem) = 13.8 (PCH_a); 3.76 dd, 1 H, *J*(P,CH_b) = 9.5 (PCH_b); 3.27 dd, 1 H, *J*(3′a,2′) = 3.0, *J*(gem) = 10.6 (H-3′a); 3.04 dd, 1 H, *J*(3′b,2′) = 3.5 (H-3′b); 1.15 d, 3 H, 1.13 d, 3 H, 1.12 d, 3 H and 1.10 d, 3 H, *J*(CH_3,CH) = 6.2 (CH_3). ^{13}C NMR (DMSO-d_6): 178.545 (C=O); 158.93 (C-4); 147.81 (C-2); 147.58 (C-6); 143.63, 3 C (Tr); 137.09 (Bz); 133.08 (Ph); 132.66 (Bz); 129.60, 2 C and 129.49, 2 C (Bz); 128.51, 2 C (Ph); 128.39, 6 C and 128.12, 6 C (Tr); 128.02, 2 C and 127.72 (Ph); 127.29, 3 C (Tr); 113.51 (C-5); 86.47 (Tr); 78.03 d, *J*(P,C) = 11.7 (C-2′); 70.32 d, 2 C, *J*(P,C) = 6.3 (P-O-C); 63.97 d, *J*(P,C) = 164.6 (P-C); 62.81 (C-3′); 50.11 (C-1′); 23.85 d, 2 C, *J*(P,C) = 3.4 and 23.70 d, 2 C, *J*(P,C) = 4.4 (CH_3).

N^4-Benzoyl-1-{(S)-2-[(diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}-5-(4-nitrophenyl)cytosine (**6b**). Chromatographed in system ethyl acetate–toluene (2:1). Yield 2.6 g (80%), yellow foam. $[\alpha]_D$ –39.2 (*c* 0.172, $CHCl_3$). For $C_{46}H_{47}N_4O_9P$ (830.9) calculated: 66.50% C, 5.70% H, 6.74% N, 3.73% P; found: 66.65% C, 5.64% H, 6.40% N, 3.87% P. FAB MS, *m/z* (%): 831 (4)

[M + H], 243 (100) [trityl]. ^{1}H NMR (DMSO-d_6): 13.50 brs, 1 H (NH); 8.32 d, 2 H (H-arom.); 8.25 s, 1 H (H-6); 8.07 d, 2 H, 7.98 d, 2 H, 7.57 t, 1 H, 7.48 t, 2 H, 7.39 d, 6 H, 7.32 t, 6 H and 7.26 t, 3 H (H-arom.); 4.51 m, 2 H (P-OCH); 4.11 m, 2 H and 4.02 m, 1 H (H-1′, H-2′); 3.84 dd, 1 H and 3.78 dd, 1 H, *J*(P,CH) = 8.8, *J*(gem) = 13.9 (PCH_2); 3.28 dd, 1 H, *J*(3′a,2′) = 3.8, *J*(gem) = 10.6 (H-3′a); 3.03 dd, 1 H, *J*(3′b,2′) = 3.2 (H-3′b); 1.14 d, 6 H and 1.12 d, 6 H, *J*(CH_3,CH) = 6.1 (CH_3). ^{13}C NMR (DMSO-d_6): 167.35 (Bz); 158.22 (C-4); 149.05 (C-6); 147.68 (C-2); 146.58, 2 C (NO_2-Ph); 143.60, 3 C (Tr); 132.78 (Bz); 130.51, 2 C (NO_2-Ph); 128.60, 4 C (Bz); 128.37, 6 C, 128.12, 6 C and 127.29, 3 C (Tr); 123.19, 2 C (NO_2-Ph); 111.24 (C-5); 86.46 (Tr); 77.93 d, *J*(P,C) = 10.2 (C-2′); 70.35 d and 70.30 d, *J*(P,C) = 6.3 (P-O-C); 63.97 d, *J*(P,C) = 164.6 (P-C); 62.82 (C-3′); 50.28 (C-1′); 23.85 d, 2 C, *J*(P,C) = 3.4, 23.80 d and 23.69 d, *J*(P,C) = 4.4 (CH_3).

N^4*-Benzoyl-5-(4-chlorophenyl)-1-{(S)-2-[(diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}-cytosine* (**6c**). Chromatographed in system ethyl acetate–toluene (1:2). Yield 2.4 g (75%), amorphous solid. $[\alpha]_D$ –40.5 (*c* 0.380, $CHCl_3$). For $C_{46}H_{47}ClN_3O_7P.3H_2O$ (874.4) calculated: 63.19% C, 6.11% H, 4.05% Cl, 4.81% N, 3.54% P; found: 63.01% C, 6.15% H, 3.98% Cl, 4.82% N, 3.43% P. FAB MS, *m/z* (%): 820 (1) [M + H], 578 (2) [M – trityl + H], 243 (68) [trityl], 105 (42) [benzoyl]. ^{1}H NMR (DMSO-d_6): 13.50 brs, 1 H (NH); 8.06 s, 1 H (H-6); 8.04 m, 2 H, 7.65 m, 2 H, 7.56 t, 1 H, 7.50 d, 2 H, 7.46 t, 2 H, 7.39 d, 6 H, 7.32 t, 6 H and 7.25 t, 3 H (H-arom.); 4.50 m, 2 H (P-OCH); 4.09 m, 2 H and 4.01 m, 1 H (H-1′, H-2′); 3.81 dd, 1 H and 3.76 dd, 1 H, *J*(P,CH) = 8.8, *J*(gem) = 13.9 (PCH_2); 3.26 dd, 1 H, *J*(3′a,2′) = 3.9, *J*(gem) = 10.5 (H-3′a); 3.02 dd, 1 H, *J*(3′b,2′) = 3.3 (H-3′b); 1.14 d, 3 H, 1.13 d, 3 H, 1.12 d, 3 H and 1.10 d, 3 H, *J*(CH_3,CH) = 6.2 (CH_3). ^{13}C NMR (DMSO-d_6): 145.70 (C-2); 143.70, 3 C (Tr); 137.54 (Cl-Ph); 132.72 (Bz); 132.32 br and 129.44 br, 4 C (Cl-Ph); 128.59, 4 C (Bz); 128.39, 6 C, 128.13, 6 C and 127.31, 3 C (Tr); 112.19 (C-5); 86.47 (Tr); 77.92 d, *J*(P,C) = 12.7 (C-2′); 70.37 d and 70.32 d, *J*(P,C) = 6.3 (P-O-C); 63.95 d, *J*(P,C) = 165.0 (P-C); 62.81 (C-3′); 23.87 d and 23.82 d, *J*(P,C) = 3.4 (CH_3); 23.70 d and 23.64 d, *J*(P,C) = 4.4 (CH_3).

Preparation of Compounds **7a**–**7c**. General Procedure

A solution of benzoylated phosphonate **6a**–**6c** (3.1 mmol) in 30% methanolic ammonia (40 ml) was kept at 4 °C for 24 h and then evaporated. The residue was chromatographed on silica gel (300 ml) in the system described below.

1-{(S)-2-[(Diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}-5-phenylcytosine (**7a**). Chromatographed in system ethyl acetate–acetone–ethanol–water (18:3:2:2). Yield 1.9 g (90%), white foam. $[\alpha]_D$ –34.8 (*c* 0.480, $CHCl_3$). For $C_{39}H_{44}N_3O_6P$ (681.8) calculated: 68.71% C, 6.50% H, 6.16% N, 4.54% P; found: 68.30% C, 6.51% H, 6.01% N, 4.83% P. FAB MS, *m/z* (%): 682 (7) [M + H], 440 (12) [M – trityl + 2 H], 243 (100) [trityl]. ^{1}H NMR (DMSO-d_6): 7.44 s, 1 H (H-6); 7.40–7.30 m, 20 H (H-arom.); 6.30 brs, 2 H (NH_2); 4.56 m, 2 H (P-OCH); 3.95 dd, 1 H, *J*(1′a,2′) = 4.0, *J*(gem) = 12.6 (H-1′a); 3.92 m, 1 H (H-2′); 3.87 dd, 1 H, *J*(1′b,2′) = 5.9 (H-1′b); 3.78 dd, 1 H, *J*(P,CH_a) = 8.9, *J*(gem) = 13.6 (PCH_a); 3.70 dd, 1 H, *J*(P,CH_b) = 9.8 (PCH_b); 3.21 dd, 1 H, *J*(3′a,2′) = 2.9, *J*(gem) = 10.6 (H-3′a); 2.93 dd, 1 H, *J*(3′b,2′) = 3.9 (H-3′b); 1.17 d, 3 H, 1.165 d, 3 H, 1.135 d, 3 H and 1.13 d, 3 H, *J*(CH_3,CH) = 6.2 (CH_3). ^{13}C NMR (DMSO-d_6): 163.795 (C-4); 155.28 (C-2); 146.36 (C-6); 143.71, 3 C (Tr); 133.96 (Ph); 129.00, 2 C and 128.83, 2 C (Ph); 128.40, 6 C and 128.09, 6 C (Tr); 127.57 (Ph); 127.24, 3 C (Tr); 106.76 (C-5); 86.255 (Tr); 78.57 d, *J*(P,C) = 13.2 (C-2′); 70.34 d, 2 C, *J*(P,C) = 6.4 (P-O-C); 64.05 d, *J*(P,C) = 166.0 (P-C); 62.86 (C-3′); 50.18 (C-1′); 23.94 d, *J*(P,C) = 3.9, 23.91 d, *J*(P,C) = 3.9, 23.83 d, *J*(P,C) = 4.4 and 23.75 d, *J*(P,C) = 4.9 (CH_3).

1-{(S)-2-[(Diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}-5-(4-nitrophenyl)cytosine (**7b**). Chromatographed in ethyl acetate–acetone–ethanol–water (15:3:4:3). Yield 2.15 g (95%), yellow foam. $[\alpha]_D$ –7.7 (*c* 0.764, $CHCl_3$). For $C_{39}H_{43}N_4O_8P$ (726.8) calculated: 64.45% C, 5.96% H, 7.71% N, 4.26% P; found: 63.88% C, 6.02% H, 7.52% N, 4.06% P. FAB MS, *m/z* (%): 749 (8) [M + Na], 243 (100) [trityl]. ^{1}H NMR (DMSO-d_6): 8.24 d, 2 H (H-arom.); 7.65 s, 1 H (H-6); 7.56 d, 2 H, 7.38 d, 6 H, 7.32 t, 6 H and 7.26 t, 3 H (H-arom); 4.52 m, 2 H (P-OCH); 3.97 dd, 1 H, *J*(1′a,2′) = 3.3, *J*(gem) = 11.8 (H-1′a); 3.93 m, 1 H (H-2′); 3.88 dd, 1 H, *J*(1′b,2′) = 7.0 (H-1′b); 3.78 dd, 1 H, *J*(P,CH_a) = 8.7, *J*(gem) = 13.7 (PCH_a); 3.70 dd, 1 H, *J*(P,CH_b) = 9.9 (PCH_b); 3.22 dd, 1 H, *J*(3′a,2′) = 2.7, *J*(gem) = 10.5 (H-3′a); 2.92 dd, 1 H, *J*(3′b,2′) = 3.9 (H-3′b); 1.17 d, 3 H, 1.16 d, 3 H, 1.135 d, 3 H, 1.13 d, 3 H, *J*(CH_3,CH) = 6.1 (CH_3). ^{13}C NMR (DMSO-d_6): 163.27 (C-4); 155.02 (C-2); 147.77 (C-6); 146.44 (NO_2-Ph); 143.70, 3 C (Tr); 141.44 and 129.88, 2 C (NO_2-Ph); 128.39, 6 C, 128.11, 6 C and 127.27, 3 C (Tr); 124.05, 2 C (NO_2-Ph); 104.94 (C-5); 86.29 (Tr). 78.49 d, *J*(P,C) = 12.7 (C-2′); 70.38 d and 70.34 d, *J*(P,C) = 6.3 (P-O-C); 64.03 d, *J*(P,C) = 165.5 (P-C); 62.83 (C-3′); 50.32 (C-1′); 24.93 d and 23.90 d, *J*(P,C) = 3.4, 23.83 d and 23.77 d, *J*(P,C) = 4.4 (CH_3).

5-(4-Chlorophenyl)-1-{(S)-2-[(diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}cytosine (**7c**). Chromatographed in ethyl acetate–acetone–ethanol–water (18:3:2:2). Yield 1.8 g (81%), white foam. $[\alpha]_D$ –29.4 (*c* 0.445, $CHCl_3$). For $C_{39}H_{43}ClN_3O_6P$ (716.2) calculated: 65.40% C, 6.05% H, 4.95% Cl, 5.87% N, 4.32% P; found: 65.27% C, 5.97% H, 4.65% Cl, 6.18% N, 4.08% P. FAB MS, *m/z* (%): 738 (10) [M + Na], 243 (100) [trityl]. ^{1}H NMR (DMSO-d_6): 7.97 br, 1 H (NH_2); 7.87 d, 2 H and 7.51 t, 1 H (H-arom.); 7.46 s, 1 H (H-6); 7.45 m, 4 H, 7.38 d, 6 H, 7.31 t, 6 H and 7.26 m, 5 H (H-arom.); 6.45 br, 1 H (NH_2); 4.55 m, 2 H (P-OCH); 3.94 dd, 1 H, *J*(1′a,2′) = 4.2, *J*(gem) = 11.8 (H-1′a); 3.90 m, 1 H (H-2′); 3.85 dd, 1 H, *J*(1′b,2′) = 6.2 (H-1′b); 3.77 dd, 1 H, *J*(P,CH_a) = 8.8, *J*(gem) = 13.7 (PCH_a); 3.69 dd, 1 H, *J*(P,CH_b) = 9.8 (PCH_b); 3.20 dd, 1 H, *J*(3′a,2′) = 3.2, *J*(gem) = 10.5 (H-3′a); 2.92 dd, 1 H, *J*(3′b,2′) = 4.0 (H-3′b); 1.17 d, 3 H, 1.165 d, 3 H, 1.133 d, 3 H and 1.131 d, 3 H, *J*(CH_3,CH) = 6.1 (CH_3). ^{13}C NMR (DMSO-d_6): 163.70 (C-4); 155.24 (C-2); 146.61 (C-6); 143.71, 3 C (Tr); 132.91, 132.25, 131.39, 2 C and 130.75, 2 C (Cl-Ph); 128.40, 6 C, 128.10, 6 C and 127.63, 3 C (Tr); 105.60 (C-5); 86.27 (Tr); 78.55 d, *J*(P,C) = 12.2 (C-2′); 70.36 d, 2 C, *J*(P,C) = 5.4 (P-O-C); 64.05 d, *J*(P,C) = 165.5 (P-C); 62.86 (C-3′); 50.21 (C-1′); 23.96 d and 23.90 d, *J*(P,C) = 3.9 (CH_3); 23.85 d and 23.76 d, *J*(P,C) = 4.4 (CH_3).

N^4-Benzoyl-1-[(*R*)-2-hydroxy-3-(trityloxy)propyl]-5-phenylcytosine (**11**)

The product was prepared from compounds **10** (2.72 g, 8.6 mmol) and **2a** (2.5 g, 8.6 mmol) by the same procedure as described for **4a**. Yield 4.51 g (86%), white foam. $[\alpha]_D$ +30.0 (*c* 0.340, $CHCl_3$). ^{1}H and ^{13}C NMR spectra are identical with those of compound **4a**.

N^4-Benzoyl-1-{(*R*)-2-[(diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}-5-phenylcytosine (**12**)

The product was prepared from **11** (4.5 g, 7.4 mmol) by the same procedure as described for **6a**. Yield 4.3 g (74%), white foam. $[\alpha]_D$ +57.2 (*c* 0.394, $CHCl_3$). ^{1}H and ^{13}C NMR spectra are identical with those of compound **6a**.

1-{(*R*)-2-[(Diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}-5-phenylcytosine (**13**)

The product was prepared from **12** (4.3 g, 5.5 mmol) by the same procedure as described for **7a**. Yield 3.1 g (82%), white foam. $[\alpha]_D$ +31.8 (*c* 0.403, $CHCl_3$). ^{1}H and ^{13}C NMR spectra are identical with those of compound **7a**.

Free Phosphonic Acids Derived from 5-Substituted Cytosines. General Procedure

Bromotrimethylsilane (2 ml, 15 mmol) was added to a solution of appropriate ester **7a**–**7c**, **14** or **26a**–**26c** (1.6 mmol) in acetonitrile (10 ml), the mixture was set aside in the dark at room temperature for 48 h and evaporated. The residue was coevaporated with acetonitrile (20 ml), dissolved in system ethanol–water–triethylamine (10:10:1, 45 ml) and after 5 min standing evaporated. The residue was partitioned between water (100 ml) and diethyl ether (100 ml), the aqueous layer concentrated to ca. 10 ml and applied onto a column of Dowex 50 (H^+ form, 50 ml). Elution was performed with water (750 ml), followed by 2.5% aqueous ammonia. UV absorbing ammonia fractions were evaporated, the residue dissolved in water (5 ml) and applied onto a column of Dowex 1 (acetate form, 50 ml). Elution was performed with water (100 ml), followed by gradient of 0–1 M acetic acid (1 l) or 1 M formic acid; concentrations necessary for product elution are given below. Product-containing fractions were evaporated, the residue coevaporated with water (4 × 20 ml) and crystallized from water or aqueous ethanol.

1-[(S)-3-Hydroxy-2-(phosphonomethoxy)propyl]-5-phenylcytosine (**8a**). Eluted from Dowex 1 with 0.3 M acetic acid. Yield 431 mg (74%), white crystals. M.p. 265–268 °C. $[\alpha]_D$ –55.0 (*c* 0.141, H_2O). For $C_{14}H_{18}N_3O_6P\cdot 0.5H_2O$ (364.3) calculated: 46.16% C, 5.26% H, 11.53% N, 8.50% P; found: 46.22% C, 5.21% H, 11.30% N, 8.39% P. FAB MS, *m/z* (%): 356 (100) [M + H], 188 (12) [5-phenylcytosine + H]. ^{1}H NMR (D_2O): 7.52 s, 1 H (H-6); 7.43 m, 3 H and 7.35 d, 2 H (H-arom.); 3.87 dd, 1 H, *J*(1′a,2′) = 6.2, *J*(gem) = 14.0 (H-1′a); 3.84 dd, 1 H, *J*(1′b,2′) = 5.8 (H-1′b); 3.71 m, 1 H (H-2′); 3.69 dd, 1 H, *J*(3′a,2′) = 3.2, *J*(gem) = 13.2 (H-3′a); 3.55 dd, 1 H, *J*(P,CH_a) = 8.3, *J*(gem) = 12.4 (PCH_a); 3.46 dd, 1 H, *J*(3′b,2′) = 6.0, *J*(gem) = 13.2 (H-3′b); 3.40 dd, 1 H, *J*(P,CH_b) = 9.5 (PCH_b). ^{13}C NMR (D_2O): 164.75 (C-4); 158.00 (C-2); 145.95 (C-6); 132.15, 129.36, 2 C, 129.24, 2 C and 128.75 (C-arom.); 109.96 (C-5); 80.11 d, *J*(P,C) = 9.8 (C-2′); 68.49 d, *J*(P,C) = 150.4 (P-C); 60.81 (C-3′); 49.965 (C-1′).

1-[(R)-3-Hydroxy-2-(phosphonomethoxy)propyl]-5-phenylcytosine (**14**). Yield 414 mg (71%), white crystals. M.p. 266 °C. $[\alpha]_D$ +54.5 (*c* 0.206, H_2O). ^{1}H and ^{13}C NMR spectra are identical with those of compound **8a**.

1-[(S)-3-Hydroxy-2-(phosphonomethoxy)propyl]-5-(4-nitrophenyl)cytosine (**8b**). Eluted from Dowex 1 with 1 M acetic acid. Yield 482 mg (72%), yellowish solid. M.p. 178 °C. $[\alpha]_D$ –13.9 (*c* 0.512, H_2O). For $C_{14}H_{17}N_4O_8P\cdot H_2O$ (418.3) calculated: 40.20% C, 4.58% H, 13.39% N, 7.40% P; found: 40.27% C, 4.55% H, 13.27% N, 7.35% P. FAB MS, *m/z* (%): 401 (12) [M + H]. ^{1}H NMR (D_2O): 8.37 d, 2 H and 7.75 d, 2 H (H-arom.); 8.04 s, 1 H (H-6); 4.26 dd, 1 H, *J*(1′a,2′) = 3.0, *J*(gem) = 14.4 (H-1′a); 3.93 dd, 1 H, *J*(1′b,2′) = 8.2 (H-1′b); 3.88 dd, 1 H, *J*(3′a,2′) = 3.9, *J*(gem) = 12.2 (H-3′a); 3.84 m, 1 H (H-2′); 3.81 dd, 1 H, *J*(P,CH_a) = J(P,CH_b) = 9.4, *J*(gem) = 13.2 (PCH_a); 3.66 dd, 1 H, *J*(3′b,2′) = 3.4 (H-3′b); 3.61 dd, 1 H (PCH_b). ^{13}C NMR (D_2O): 158.49 (C-4); 149.99 (C-6); 148.99 (C-2); 148.13, 136.24, 130.97, 2 C and 124.52, 2 C (C-arom.); 106.51 (C-5); 79.06 d, *J*(P,C) = 11.2 (C-2′); 65.87 d, *J*(P,C) = 157.7 (P-C); 59.82 (C-3′); 50.60 (C-1′).

5-(4-Chlorophenyl)-1-[(S)-3-hydroxy-2-(phosphonomethoxy)propyl]cytosine (**8c**). Eluted from Dowex 1 with 0.5 M acetic acid. Yield 525 mg (82%), white crystals. M.p. 293 °C. $[\alpha]_D$ –37.2

(*c* 0.329, 0.1 M HCl). For $C_{14}H_{17}ClN_3O_6P \cdot 0.5H_2O$ (398.7) calculated: 42.17% C, 4.54% H, 8.90% Cl, 10.53% N, 7.77% P; found: 42.31% C, 4.31% H, 9.10% Cl, 10.27% N, 7.44% P. FAB MS, *m/z* (%): 390 (100) [M + H]. ^{1}H NMR (D_2O): 7.63 s, 1 H (H-6); 7.51 d, 2 H and 7.39 d, 2 H (H-arom.); 3.95 d, 2 H, *J*(1′,2′) = 5.9 (H-1′); 3.78 m, 1 H (H-2′); 3.78 dd, 1 H, *J*(3′a,2′) = 2.8, *J*(gem) = 13.2 (H-3′a); 3.62 dd, 1 H, *J*(P,CH_a) = 8.4, *J*(gem) = 12.4 (PCH_a); 3.55 dd, 1 H, *J*(3′b,2′) = 6.1 (H-3′b); 3.48 dd, 1 H, *J*(P,CH_b) = 9.5 (PCH_b). ^{13}C NMR (D_2O): 164.60 (C-4); 157.95 (C-2); 146.12 (C-6); 133.93, 130.83, 2 C, 130.79 and 129.24, 2 C (C-arom.); 108.92 (C-5); 80.14 d, *J*(P,C) = 9.8 (C-2′); 68.46 d, *J*(P,C) = 150.4 (P-C); 60.77 (C-3′); 50.01 (C-1′).

5-Benzyl-1-[(S)-3-hydroxy-2-(phosphonomethoxy)propyl]cytosine (**25a**). Eluted from Dowex 1 with 0.5 M acetic acid. Yield 421 mg (68%), white solid. $[\alpha]_D$ –32.5 (*c* 0.412, H_2O). For $C_{15}H_{20}N_3O_6P \cdot H_2O$ (387.3) calculated: 46.52% C, 5.72% H, 10.85% N, 8.00% P; found: 46.53% C, 5.72% H, 10.80% N, 7.75% P. ESI MS, *m/z* (%): 370.1 (50) [M + H], 738.9 (100) [2 M + H]. ^{1}H NMR (D_2O): 7.65 s, 1 H (H-6); 7.43 m, 2 H and 7.33 m, 3 H (H-arom.); 4.11 dd, 1 H, *J*(1′a,2′) = 3.4, *J*(gem) = 14.2 (H-1′a); 3.86 dd, 1 H, *J*(1′b,2′) = 7.8 (H-1′b); 3.84 s, 2 H (CH_2); 3.82 dd, 1 H, *J*(3′a,2′) = 3.9, *J*(gem) = 11.7 (H-3′a); 3.79 m, 1 H (H-2′); 3.74 dd, 1 H and 3.59 dd, 1 H, *J*(P,CH) = 9.2, *J*(gem) = 13.2 (PCH_2); 3.59 dd, 1 H, *J*(3′b,2′) = 3.2 (H-3′b). ^{13}C NMR (D_2O): 158.84 (C-4); 149.36 (C-6); 148.98 (C-2); 135.96, 129.03, 2 C, 128.51, 2 C and 127.28 (Ph); 105.53 (C-5); 79.17 d, *J*(P,C) = 10.7 (C-2′); 65.86 d, *J*(P,C) = 157.7 (P-C); 59.94 (C-3′); 49.94 (C-1′); 31.12 (CH_2-Ph).

1-[(S)-3-Hydroxy-2-(phosphonomethoxy)propyl]-5-(1-naphthylmethyl)cytosine (**25b**). Eluted from Dowex 1 with 1 M formic acid. Yield 389 mg (58%), white solid. $[\alpha]_D$ –40.4 (*c* 0.136, CH_3OH + 0.1 M HCl, 10:1). FAB MS, *m/z* (%): 420 (100) [M + H]. HR MS (FAB): For $C_{19}H_{23}N_3O_6P$ [M + H] calculated: 420.1324; found: 420.1329. ^{1}H NMR (D_2O): 7.95 d, 1 H, 7.86 m, 2 H, 7.56 m, 2 H, 7.47 t, 1 H, 7.30 d and 1 H (H-arom.); 6.79 s, 1 H (H-6); 3.99 m, 2 H (CH_2); 3.60 dd, 1 H, *J*(1′a,2′) = 5.3, *J*(gem) = 12.9 (H-1′a); 3.43–3.55 m, 4 H (H-1′b, H-2′, H-3′a, OCH_2P_a); 3.36 dd, 1 H, *J*(P,CH_b) = 9.2, *J*(gem) = 12.7 (PCH_b); 3.23 dd, 1 H, *J*(3′b,2′) = 5.0, *J*(gem) = 12.4 (H-3′b). ^{13}C NMR (D_2O): 165.95 (C-4); 158.51 (C-2); 146.47 (C-6); 134.22, 133.33, 131.94, 129.47, 128.42, 127.42, 127.34, 126.89, 126.57 and 124.22 (1-naphthyl); 106.93 (C-5); 80.24 d, *J*(C,P) = 10.0 (C-2′); 68.22 d, *J*(C,P) = 153.0 (P-C); 61.12 (C-3′); 50.10 (C-1′); 30.24 (CH_2-naphthyl).

1-[(S)-3-Hydroxy-2-(phosphonomethoxy)propyl]-5-(2-naphthylmethyl)cytosine (**25c**). Eluted from Dowex 1 with 1 M formic acid. Yield 389 mg (58%), white solid. $[\alpha]_D$ not determined (opalescent solutions in H_2O, CH_3OH or 0.1 M NaOH). FAB MS, *m/z* (%): 420 (100) [M + H]. HR MS (FAB): For $C_{19}H_{23}N_3O_6P$ [M + H] calculated: 420.1324; found: 420.1345. ^{1}H NMR (D_2O): 7.90–7.79 m, 4 H (H-arom.); 7.77 s, 1 H (H-6); 7.52–7.41 m, 3 H (H-arom.); 4.05 m, 1 H (H-1′a); 3.88 s, 2 H (CH_2); 3.75–3.51 m, 4 H (H-1′b, H-2′, PCH_2); 3.45 m, 2 H (H-3′). ^{13}C NMR (D_2O): 161.01 (C-4); 150.64 (C-2); 149.04 (C-6); 135.72, 133.24, 132.04, 128.08, 127.71, 127.68, 127.49, 126.68, 126.31 and 125.81 (2-naphthyl); 103.89 (C-5); 79.72 d, *J*(C,P) = 8.5 (C-2′); 65.88 d, *J*(C,P) = 159.0 (P-C); 60.58 (C-3′); 49.70 (C-1′); 31.39 (CH_2-naphthyl).

5-(4-Chlorophenyl)-1-[(*S*)-3-hydroxy-2-(phosphonomethoxy)propyl]cytosine (**8d**)

A solution of **4d** (447 mg, 0.7 mmol) and **5** (350 mg, 1 mmol) in DMF (25 ml) was cooled to –20 °C and 60% oil dispersion of NaH (84 mg, 2.1 mmol) was added. The mixture was stirred at –20 °C for 30 min, then left to warm to room temperature and the stirring continued for additional 16 h. The solution was neutralized with acetic acid to pH 7, evaporated and the residue coevaporated with toluene. 30% methanolic ammonia (20 ml) was added,

the resulting solution kept at 4 °C for 24 h and evaporated. The crude product **7d** was deprotected with bromotrimethylsilane by the same procedure as described for compounds **8a**–**8c**. The product **8d** was eluted from Dowex 1 with 0.3 M acetic acid. Yield 190 mg (70%), white amorphous solid. $[\alpha]_D$ –28.6 (*c* 0.219, H_2O). FAB MS, *m/z* (%): 390 (34) [M + H]. HR MS (FAB): For $C_{14}H_{18}ClN_3O_6P$ [M + H] calculated: 390.0622; found: 390.0615. ^{1}H NMR (D_2O + NaOD): 7.64 s, 1 H (H-6); 7.60 m, 1 H and 7.47 m, 3 H (H-arom.); 3.98 br dd, 1 H, *J*(1′a,2′) = 3.0, *J*(gem) = 13.9 (H-1′a); 3.91 dd, 1 H, *J*(1′b,2′) = 5.7 (H-1′b); 3.80 m, 1 H (H-2′); 3.76 dd, 1 H, *J*(3′a,2′) = 3.0, *J*(gem) = 12.6 (H-3′a); 3.65 br dd, 1 H and 3.59 br dd, 1 H, *J*(P,CH) = 9.0, *J*(gem) = 13.2 (PCH_2); 3.56 dd, 1 H, *J*(3′b,2′) = 5.9 (H-3′b). ^{13}C NMR (D_2O + NaOD): 164.61 (C-4); 158.07 (C-2); 146.70 (C-6); 134.26, 132.59, 130.89, 130.46, 129.87 and 127.80 (C-arom.); 107.45 (C-5); 80.60 d, *J*(P,C) = 10.7 (C-2′); 68.59 d, *J*(P,C) = 150.4 (P-C); 60.95 (C-3′); 49.85 (C-1′).

1-[(*S*)-3-Hydroxy-2-(phosphonomethoxy)propyl]-5-phenyluracils (**9a**–**9d**, **15**). General Procedure

A solution of appropriate cytosine phosphonate **8a**–**8d** or **14** (0.28 mmol) in 80% acetic acid (3.5 ml) was kept with 3-methylbutyl nitrite (0.5 ml, 3.7 mmol) at room temperature for 3 days. The solution was evaporated, the residue coevaporated with water (2 × 5 ml) and applied onto a column of Dowex 50 (H^+, 25 ml). The column was eluted with water, the UV-absorbing eluate adjusted to pH 3 and deionized on activated charcoal. The final purification was performed on preparative reverse phase HPLC column, isocratically in aqueous methanol (concentration given bellow), flow rate 12 ml/min. The pure products were lyophilized from water, some of them transformed to dilithium salts before lyophilization.

1-[(S)-3-Hydroxy-2-(phosphonomethoxy)propyl]-5-phenyluracil (**9a**). Yield 50 mg (49%) of dilithium salt, white amorphous solid. M.p. 180–185 °C. Elution from HPLC column: 10% CH_3OH. $[\alpha]_D$ –18.2 (*c* 0.233, H_2O). For $C_{14}H_{15}Li_2N_2O_7P$ (368.1) calculated: 45.68% C, 4.11% H, 7.61% N, 8.41% P; found: 45.34% C, 4.63% H, 7.71% N, 8.33% P. FAB MS, *m/z* (%): 369 (0.5) [M + H]. ^{1}H NMR (D_2O): 7.73 s, 1 H (H-6); 7.46 m, 5 H (H-arom.); 4.03 dd, 1 H, *J*(1′a,2′) = 4.5, *J*(gem) = 14.4 (H-1′a); 3.89 dd, 1 H, *J*(1′b,2′) = 7.2 (H-1′b); 3.84 dd, 1 H, *J*(3′a,2′) = 3.7, *J*(gem) = 12.4 (H-3′a); 3.79 m, 1 H (H-2′); 3.72 dd, 1 H, *J*(P,CH_a) = 9.6, *J*(gem) = 12.9 (PCH_a); 3.67 dd, 1 H, *J*(P,CH_b) = 9.2 (PCH_b); 3.62 dd, 1 H, *J*(3′b,2′) = 4.4 (H-3′b). ^{13}C NMR (D_2O): 165.09 (C-4); 151.97 (C-2); 145.13 (C-6); 131.81, 128.63, 2 C, 128.56, 2 C and 128.19 (C-arom.); 114.79 (C-5); 79.75 d, *J*(P,C) = 11.7 (C-2′); 66.51 d, *J*(C,P) = 156.2 (P-C); 60.27 (C-3′); 49.07 (C-1′).

1-[(R)-3-Hydroxy-2-(phosphonomethoxy)propyl]-5-phenyluracil (**15**). Yield 61 mg (59%) of dilithium salt. $[\alpha]_D$ +20.0 (*c* 0.459, H_2O). M.p. 181–187 °C. MS and NMR data are identical with those of compound **9a**.

1-[(S)-3-Hydroxy-2-(phosphonomethoxy)propyl]-5-(4-nitrophenyl)uracil (**9b**). Yield 49 mg (42%) of dilithium salt, yellow amorphous solid. $[\alpha]_D$ +11.64 (*c* 0.272, H_2O). Elution from HPLC column: 10% CH_3OH. HR MS (FAB): For $C_{14}H_{15}Li_2N_3O_9P$ [M + H] calculated: 414.0866; found: 414.0878. FAB MS, *m/z* (%): 414 (2) [M + H]. ^{1}H NMR (D_2O): 8.29 d, 2 H (H-arom.); 8.02 s, 1 H (H-6); 7.77 d, 2 H (H-arom.); 4.08 dd, 1 H, *J*(1′a,2′) = 4.4, *J*(gem) = 14.4 (H-1′a); 3.99 dd, 1 H, *J*(1′b,2′) = 7.0 (H-1′b); 3.86 dd, 1 H, *J*(3′a,2′) = 3.0, *J*(gem) = 12.2 (H-3′a); 3.80 m, 1 H (H-2′); 3.65 dd, 1 H, *J*(P,CH_a) = 9.4, *J*(gem) = 12.4 (PCH_a); 3.61 dd, 1 H, *J*(3′b,2′) = 4.4 (H-3′b); 3.60 dd, 1 H, *J*(P,CH_b) = 9.4 (PCH_b). ^{13}C NMR (D_2O): 164.55 (C-4); 151.98 (C-2);

146.82 (C-arom.); 146.54 (C-6); 139.19, 129.30, 2 C and 123.72, 2 C (C-arom.); 79.62 d, *J*(P,C) = 10.2 (C-2′); 65.50 d, *J*(P,C) = 156.0 (P-C); 60.41 (C-3′); 49.23 (C-1′).

5-(4-Chlorophenyl)-1-[(S)-3-hydroxy-2-(phosphonomethoxy)propyl]uracil (**9c**). Yield 44 mg (40%) of free acid, white amorphous solid. $[\alpha]_D$ –3.8 (*c* 0.325, H_2O). Elution from HPLC column: 15% CH_3OH. FAB MS, *m/z* (%): 391 (15) [M + H]. HR MS (FAB): For $C_{14}H_{17}ClN_2O_7P$ [M + H] calculated: 391.0462; found: 391.0451. ^{1}H NMR (D_2O): 7.75 s, 1 H (H-6); 7.47 m, 4 H (H-arom.); 4.07 dd, 1 H, *J*(1′a,2′) = 3.8, *J*(gem) = 14.6 (H-1′a); 3.89 dd, 1 H, *J*(1′b,2′) = 7.7 (H-1′b); 3.85 dd, 1 H, *J*(3′a,2′) = 3.7, *J*(gem) = 12.7 (H-3′a); 3.79 m, 1 H (H-2′); 3.75 dd, 1 H, *J*(P,CH_a) = 4.7, *J*(gem) = 13.0 (PCH_a); 3.66 dd, 1 H, *J*(P,CH_b) = 9.2 (PCH_b); 3.63 dd, 1 H, *J*(3′b,2′) = 4.1 (H-3′b). ^{13}C NMR (D_2O): 164.88 (C-4); 151.90 (C-2); 145.29 (C-6); 133.35, 130.43, 130.01, 2 C and 128.51, 2 C (C-arom.); 113.66 (C-5); 79.76 d, *J*(P,C) = 12.2 (C-2′); 66.22 d, *J*(P,C) = 158.2 (P-C); 60.18 (C-3′); 49.20 (C-1′).

5-(2-Chlorophenyl)-1-[(S)-3-hydroxy-2-(phosphonomethoxy)propyl]uracil (**9d**). Yield 90 mg (76%) of monoammonium salt, white solid. $[\alpha]_D$ –24.7 (*c* 0.120, H_2O). Elution from HPLC column: 15% CH_3OH. FAB MS, *m/z* (%): 413 (8) [M + Na] of free acid. For $C_{14}H_{19}ClN_3O_7P{\cdot}H_2O$ (425.8) calculated: 39.49% C, 4.97% H, 8.33% Cl, 9.87% N, 7.27% P; found: 39.19% C, 4.68% H, 8.18% Cl, 9.73% N, 6.90% P. ^{1}H NMR (D_2O): 7.78 s, 1 H (H-6); 7.56 m, 1 H and 7.43 m, 3 H (H-arom.); 4.09 dd, 1 H, *J*(1′a,2′) = 4.0, *J*(gem) = 14.4 (H-1′a); 3.92 dd, 1 H, *J*(1′b,2′) = 7.2 (H-1′b); 3.85 dd, 1 H, *J*(3′a,2′) = 3.8, *J*(gem) = 12.1 (H-3′a); 3.82 m, 1 H (H-2′); 3.77 dd, 1 H and 3.71 dd, 1 H, *J*(P,CH) = 9.6, *J*(gem) = 13.2 (PCH_2); 3.64 dd, 1 H, *J*(3′b,2′) = 3.9 (H-3′b). ^{13}C NMR (D_2O): 164.915 (C-4); 152.19 (C-2); 146.79 (C-6); 134.13, 132.28, 130.45, 130.39, 129.42 and 127.29 (C-arom.); 113.34 (C-5); 79.79 d, *J*(P,C) = 11.7 (C-2′); 66.33 d, *J*(P,C) = 157.2 (P-C); 60.25 (C-3′); 49.12 (C-1′).

5-Phenyl-1-[(*R*)-2-(phosphonomethoxy)propyl]cytosine (**18**)

The synthon **16** (530 mg, 1.34 mmol) was added in several portions during 5 h to a mixture of **2a** (437 mg, 1.5 mmol) and cesium carbonate (85 mg, 0.25 mmol) in DMF (5 ml) stirred at 120 °C. Additional portion of **16** (530 mg, 1.34 mmol) was added and the heating continued for 20 h. The mixture was evaporated, the residue coevaporated with toluene (2 × 10 ml), then set aside for 48 h with 30% methanolic ammonia (20 ml) at 4 °C and evaporated. The residue was stirred in acetonitrile (20 ml) with bromotrimethylsilane (3 ml, 22.2 mmol) at room temperature for 24 h, evaporated and coevaporated with acetonitrile (5 ml). The mixture of ethanol, water and triethylamine (10, 10 and 2 ml) was added and the solution after 5 min evaporated. The residue was dissolved in water (5 ml) and desalted on Dowex 50, followed by purification on Dowex 1 (the same procedure as described for compounds **8a**–**8d**). Elution from Dowex 1 was performed with 0.3 M acetic acid. Yield 234 mg (46%) of white solid. M.p. 289–293 °C. HR MS (FAB): For $C_{14}H_{19}N_3O_5P$ [M + H] calculated: 340.1062; found: 340.1067. FAB MS, *m/z* (%): 340 (100) [M + H]. ^{1}H NMR (D_2O + NaOD): 7.63 s, 1 H (H-6); 7.52 m, 2 H and 7.43 m, 3 H (H-arom.); 4.00 dd, 1 H, *J*(1′a,2′) = 6.0, *J*(gem) = 13.6 (H-1′a); 3.92 m, 1 H (H-2′); 3.84 dd, 1 H, *J*(1′b,2′) = 5.2 (H-1′b); 3.84 dd, 1 H, *J*(PCH_a) = 8.9, *J*(gem) = 12.6 (PCH_a); 3.52 dd, 1 H, *J*(P,CH_b) = 9.3 (PCH_b); 1.19 d, 3 H, *J*(3′,2′) = 6.2 (CH_3). ^{13}C NMR (D_2O + NaOD): 164.73 (C-4); 158.16 (C-2); 146.12 (C-6); 132.23, 129.37, 2 C, 129.32, 2 C and 128.49 (C-arom.); 75.54 d, *J*(P,C) = 10.3 (C-2′); 67.28 d, *J*(P,C) = 152.3 (P-C); 53.54 (C-1′); 16.67 (C-3′).

5-Phenyl-1-[(*R*)-2-(phosphonomethoxy)propyl]uracil (**19**)

The compound was prepared from cytosine derivative **18** (150 mg, 0.44 mmol) by the same procedure as described for **9a**–**9d**. Yield 50 mg (33%) of white amorphous solid. $[\alpha]_D$ +16.1 (*c* 0.222, H_2O). Elution from HPLC column: 10% CH_3OH. FAB MS, *m/z* (%): 341 (44) [M + H]. HR MS (FAB): For $C_{14}H_{18}N_2O_6P$ [M + H] calculated: 341.0903; found: 341.0914. ^{1}H NMR (D_2O): 7.75 s, 1 H (H-6); 7.51 m, 4 H and 7.45 m, 1 H (H-arom.); 3.96 dd, 1 H, *J*(1′a,2′) = 3.4, *J*(gem) = 13.9 (H-1′a); 3.90 m, 1 H (H-2′); 3.82 dd, 1 H, *J*(1′b,2′) = 7.2 (H-1′b); 3.71 dd, 1 H and 3.56 dd, 1 H, *J*(P,CH) = 9.3, *J*(gem) = 13.2 (PCH_2); 1.23 d, 3 H, *J*(3′,2′) = 6.2 (H-3′). ^{13}C NMR (D_2O): 165.18 (C-4); 152.10 (C-2); 145.30 (C-6); 114.69 (C-5); 75.89 d, *J*(P,C) = 11.2 (C-2′); 65.00 d, *J*(P,C) = 158.7 (P-C); 52.59 (C-1′); 16.1 (C-3′).

Transformation of 5-Substituted Uracils to 4-Methoxy Derivatives **21a**–**21c**

A suspension of a uracil derivative **20a**–**20c** (10.45 mmol) in $POCl_3$ (6 ml, 64.5 mmol) was heated with *N*,*N*-dimethylaniline (2 ml, 15.7 mmol) at 110 °C for 3 h (till dissolution). $POCl_3$ was distilled off, the liquid residue poured onto ice and the product (dichloro derivative) extracted with ether (200 ml). The organic layer was extracted with cold water (100 ml), followed by saturated solution of sodium hydrogencarbonate (2 × 100 ml), dried with anhydrous sodium sulfate and evaporated. The residue was dissolved in methanol (5 ml), 1 M sodium methoxide (20 ml) was added, the mixture refluxed for 8 h and then set aside at room temperature for 12 h. The precipitated NaCl was filtered off, washed with methanol and combined filtrates were evaporated. The residue was partitioned between water (40 ml) and ether (3 × 40 ml), the combined ether extracts dried with anhydrous magnesium sulfate and evaporated. The liquid residue was stirred with acetyl chloride (6 ml) at room temperature for 24 h. Acetyl chloride was distilled off under reduced pressure, the residue codistilled with toluene (2 × 20 ml) and then dissolved in methanol (5 ml). 1 M sodium methoxide was added until the solution was alkaline, the mixture was heated at 40 °C for 1 h, then neutralized with acetic acid to pH 7 and evaporated. A mixture of acetone–ether (1:1) was added, the white crystalline material filtered by suction, washed successively with acetone, water, acetone and finally with a mixture acetone–ether (1:1) and dried in vacuo.

5-Benzyl-4-methoxypyrimidin-2(1H)-one (**21a**). Yield 1.7 g (76%), white crystals. M.p. 207–209 °C. FAB MS, *m/z* (%): 217 (100) [M + H]. For $C_{12}H_{12}N_2O_2$ (216.2) calculated: 66.65% C, 5.59% H, 12.95% N; found: 66.15% C, 5.59% H, 12.59% N. ^{1}H NMR (DMSO-d_6): 11.20 brs, 1 H (NH); 7.51 s, 1 H (H-6); 7.30 m, 2 H and 7.20 m, 3 H (H-arom.); 3.78 s, 3 H (OCH_3); 3.60 s, 2 H (CH_2). ^{13}C NMR (DMSO-d_6): 170.46 (C-4); 156.34 (C-2); 143.42 (C-6); 139.68, 128.53, 4 C and 126.31 (C-arom.); 106.02 (C-5); 53.90 (OCH_3); 31.62 (CH_2).

4-Methoxy-5-(1-naphthylmethyl)pyrimidin-2(1H)-one (**21b**). Yield 1.6 g (56%), white crystals. M.p. 231 °C. FAB MS, *m/z* (%): 267 (100) [M + H]. For $C_{16}H_{14}N_2O_2 \cdot 0.25H_2O$ (270.8) calculated: 70.97% C, 5.40% H, 10.34% N; found: 71.01% C, 5.53% H, 10.01% N. ^{1}H NMR (DMSO-d_6): 11.15 brs, 1 H (NH); 8.04 d, 1 H, 7.93 d, 1 H, 7.82 d, 1 H, 7.54 m, 2 H, 7.44 t, 1 H and 7.27 d, 1 H (H-arom.); 7.18 s, 1 H (H-6); 4.07 s, 2 H (CH_2); 3.82 s, 3 H (OCH_3). ^{13}C NMR (DMSO-d_6): 170.52 (C-4); 156.29 (C-2); 143.43 (C-6); 134.90, 133.59, 131.48, 128.82, 127.22, 126.49, 126.47, 125.97, 125.86 and 123.85 (C-arom.); 105.21 (C-5); 54.02 (OCH_3); 28.57 (CH_2).

4-Methoxy-5-(2-naphthylmethyl)pyrimidin-2(1H)-one (**21c**). Yield 2.2 g (78%), white crystals. M.p. 220–223 °C. FAB MS, *m/z* (%): 267 (20) [M + H]. For $C_{16}H_{14}N_2O_2 \cdot 0.25H_2O$ (270.8) calculated: 70.97% C, 5.40% H, 10.34% N; found: 71.03% C, 5.29% H, 10.29% N. ^{1}H NMR

(DMSO-d_6): 11.25 brs, 1 H (NH); 7.87–7.82 m, 3 H (H-arom.); 7.66 m, 1 H (H-arom.); 7.59 s, 1 H (H-6); 7.49–7.43 m, 2 H and 7.38 dd, 1 H (H-arom.); 3.78 s, 5 H (OCH_3, CH_2). ^{13}C NMR (DMSO-d_6): 170.50 (C-4); 156.36 (C-2); 143.65 (C-4); 137.33, 133.25, 131.86, 128.00, 127.63, 127.58, 127.34, 126.44, 126.24 and 126.61 (C-arom.); 105.76 (C-5); 53.91 (OCH_3); 31.78 (CH_2).

Reaction of (2*S*)-2-[(Trityloxy)methyl]oxirane (**3**) with Compounds **21a**–**21c**. General Procedure. Method *A*

Compound **3** (2.1 g, 6.7 mmol) and cesium carbonate (220 mg, 0.68 mmol) were added to a stirred suspension of 4-methoxy derivative **21** (6.3 mmol) in DMF (60 ml). The mixture was heated at 80 °C until the conversion was complete (10–12 h, TLC check). The mixture was cooled to room temperature, evaporated and the residue coevaporated with toluene (2 × 50 ml). The crude product was purified by chromatography on silica gel (750 ml) in system toluene–ethyl acetate (1:1) containing 1% triethylamine.

5-Benzyl-1-[(S)-2-hydroxy-3-(trityloxy)propyl]-4-methoxypyrimidin-2(1H)-one (**22a**). Yield 2 g (60%), white foam. $[\alpha]_D$ –16.5 (*c* 0.427, $CHCl_3$). ESI MS, *m/z* (%): 555 (100) [M + Na], 1087 (86) [2 M – H + Na], 1088 (50) [2 M + Na]. For $C_{34}H_{32}N_2O_4$ (532.7) calculated: 76.67% C, 6.06% H, 5.26% N; found: 76.63% C, 6.11% H, 5.08% N. ^{1}H NMR (DMSO-d_6): 7.70 s, 1 H (H-6); 7.41 d, 6 H, 7.33 t, 6 H, 7.26 m, 5 H and 7.18 t, 3 H (H-arom.); 5.31d, 1 H, *J*(OH,2′) = 5.7 (OH); 4.09 dd, 1 H, *J*(1′a,2′) = 3.7, *J*(gem) = 13.2 (H-1′a); 3.99 m, 1 H (H-2′); 3.78 s, 3 H (OCH_3); 3.60 d, 1 H and 3.56 d, 1 H, *J*(gem) = 15.4 (CH_2); 3.53 dd, 1 H, *J*(1′b,2′) = 8.8 (H-1′b); 2.99 dd, 1 H, *J*(3′a,2′) = 5.3, *J*(gem) = 9.4 (H-3′a); 2.90 dd, 1 H, *J*(3′b,2′) = 5.3 (H-3′b). ^{13}C NMR (DMSO-d_6): 169.715 (C-4); 155.54 (C-2); 148.54 (C-6); 143.91, 3 C (Tr); 139.61 and 128.48, 2 C (Ph); 128.45, 6 C (Tr); 128.38, 2 C (Ph); 128.06, 6 C and 127.19, 3 C (Tr); 126.95 (Ph); 105.41 (C-5); 86.06 (Tr); 67.07 (C-2′); 66.30 (C-3′); 54.06 (OCH_3); 53.28 (C-1′); 31.73 (CH_2).

1-[(S)-2-Hydroxy-3-(trityloxy)propyl]-4-methoxy-5-(1-naphthylmethyl)pyrimidin-2(1H)-one (**22b**). Yield 2.5 g (67%), white crystals. M.p. 150 °C (toluene). $[\alpha]_D$ –27.6 (*c* 0.243, $CHCl_3$). ESI MS, *m/z* (%): 605 (100) [M + Na], 606 (36) [M + H + Na]. For $C_{38}H_{34}N_2O_4$ (582.7) calculated: 78.33% C, 5.88% H, 4.81% N; found: 77.94% C, 5.76% H, 4.59% N. ^{1}H NMR (DMSO-d_6): 8.08 d, 1 H, 7.93 d, 1 H and 7.80 d, 1 H (H-arom); 7.64 s, 1 H (H-6); 7.54 m, 2 H, 7.41 t, 1 H, 7.39 d, 6 H, 7.31 t, 6 H, 7.25 t, 3 H and 7.18 d, 1 H (H-arom.); 5.23 d, 1 H, *J*(OH,2′) = 5.9 (OH); 4.07 dd, 1 H, *J*(1′a,2′) = 3.4, *J*(gem) = 13.0 (H-1′a); 4.06 s, 2 H (CH_2); 3.97 m, 1 H (H-2′); 3.79 s, 3 H (OCH_3); 3.47 dd, 1 H, *J*(1′b,2′) = 8.8 (H-1′b); 2.97 dd, 1 H and 2.86 dd, 1 H, *J*(3′,2′) = 5.4, *J*(gem) = 9.2 (H-3′). ^{13}C NMR (DMSO-d_6): 169.84 (C-4); 155.56 (C-2); 148.86 (C-6); 143.92, 3 C (Tr); 135.02, 133.52, 131.51 and 128.74 (naphthyl); 128.44, 6 C, 128.05, 6 C and 127.19, 3 C (Tr); 127.09, 126.40, 125.98, 125.92, 125.75 and 123.76 (1-naphthyl); 104.50 (C-5); 86.06 (Tr); 67.08 (C-2′); 66.23 (C-3′); 54.12 (OCH_3); 53.29 (C-1′); 28.57 (CH_2).

1-[(S)-2-Hydroxy-3-(trityloxy)propyl]-4-methoxy-5-(2-naphthylmethyl)pyrimidin-2(1H)-one (**22c**). Yield 2.6 g (72%), white foam. $[\alpha]_D$ –25.0 (*c* 0.220, $CHCl_3$). FAB MS, *m/z* (%): 605 (17) [M + Na], 583 (2) [M + H], 243 (100) [trityl]. For $C_{38}H_{34}N_2O_4$ (582.7) calculated: 78.33% C, 5.88% H, 4.81% N; found: 78.25% C, 5.77% H, 4.67% N. ^{1}H NMR (DMSO-d_6): 7.88–7.80 m, 4 H (H-6, H-arom.); 7.65 m, 1 H, 7.49–7.43 m, 2 H, 7.41 m, 6 H, 7.36 dd, 1 H, 7.33 m, 6 H and 7.25 m, 3 H (H-arom.); 5.32 d, 1 H, *J*(OH,2′) = 5.8 (OH); 4.12 dd, 1 H, *J*(1′a,2′) = 3.5, *J*(gem) = 13.1 (H-1′a); 4.02 m, 1 H (H-2′); 3.78 s, 3 H (OCH_3); 3.76 m, 2 H (CH_2); 3.55 dd, 1 H, *J*(1′b,2′) = 8.8, *J*(gem) = 13.1 (H-1′b); 3.01 dd, 1 H, *J*(3′a,2′) = 5.2, *J*(gem) = 9.4 (H-3′a);

2.91 dd, 1 H, *J*(3′b,2′) = 5.4. ^{13}C NMR (DMSO-d_6): 169.79 (C-4); 155.59 (C-2); 148.78 (C-6); 143.92 (Tr); 137.24, 133.26 and 131.91 (2-naphthyl); 128.46 and 128.07 (Tr); 127.98, 127.67, 127.62 and 127.30 (2-naphthyl); 127.20 (Tr); 126.44, 126.24 and 125.63 (naphthyl); 105.23 (C-5); 86.08 (Tr); 67.08 (C-2′); 66.33 (C-3′); 54.11 (OCH_3); 53.36 (C-1′); 31.88 (CH_2).

Reaction of (2*S*)-2-[(Trityloxy)methyl]oxirane (**3**) with **21a**. Method *B*

The same procedure as described for method *A* but the reaction was performed at 110 °C for 4 h. The chromatography afforded four fractions in the following elution order: **28**, **27**, **22a** (1.5 g, 45%) and 5-benzyluracil (153 mg, 12%).

5-Benzyl-1,3-bis[(2S)-2-hydroxy-3-(trityloxy)propyl]uracil (**28**). Yield 421 mg (8%), white foam. $[\alpha]_D$ –12.4 (*c* 0.349, $CHCl_3$). ESI MS, *m/z* (%): 857 (100) [M – H + Na], 858 (55) [M + Na]. For $C_{55}H_{50}N_2O_6$ (835.0) calculated: 79.11% C, 6.04% H, 3.35% N; found: 78.95% C, 5.99% H, 3.28% N. ^{1}H NMR (DMSO-d_6): 7.49 s, 1 H (H-6); 7.40–7.15 m, 35 H (H-arom.); 5.26 d, 1 H, *J* = 5.6 and 5.00 d, 1 H, *J* = 5.8 (OH); 3.87 dd, 1 H, *J* = 5.0 and 13.2, 3.76 dd, 1 H, *J* = 6.1 and 13.2, 3.37 dd, 1 H, *J* = 8.8 and 13.2 and 3.34 dd, 1 H, *J* = 8.3 and 13.2 (H-1′); 3.98 m, 1 H and 3.88 m, 1 H (H-2′); 3.88 d, 2 H and 3.85 d, 2 H, *J*(gem) = 15.6 (CH_2); 3.00 dd, 1 H, *J* = 5.5 and 9.4, 2.99 dd, 1 H, *J* = 4.8 and 9.4, 2.86 dd, 1 H, *J* = 5.6 and 9.4 and 2.80 dd, 1 H, *J* = 5.4 and 9.4 (H-3′). ^{13}C NMR (DMSO-d_6): 162.97 (C-4); 151.17 (C-2); 144.00 and 143.87 (Tr); 142.70 (C-6); 139.92 and 128.56, 2 C (Ph); 128.42, 12 C (Tr); 128.33, 2 C (Ph); 128.06, 6 C, 127.93, 6 C, 127.20, 3 C and 127.08, 3 C (Tr); 126.15 (Ph); 110.57 (C-5); 86.12 and 86.11 (Tr); 67.37 and 66.78 (C-2′); 67.27 and 66.14 (C-3′); 52.74 and 44.53 (C-1′); 32.65 (CH_2).

5-Benzyl-1-[(S)-2-hydroxy-3-(trityloxy)propyl]uracil (**27**). Yield 849 mg (26%), white foam. $[\alpha]_D$ –27.4 (*c* 0.460, $CHCl_3$). For $C_{33}H_{30}N_2O_4$ (518.6) calculated: 76.43% C, 5.83% H, 5.40% N; found: 76.89% C, 6.05% H, 5.01% N. ^{1}H NMR (DMSO-d_6): 7.57 s, 1 H (H-6); 7.40 d, 6 H, 7.32 t, 6 H and 7.28–7.14 m, 8 H (H-arom.); 5.31 d, 1 H, *J*(OH,2′) = 5.4 (OH); 4.03 dd, 1 H, *J*(1′a,2′) = 3.9, *J*(gem) = 13.4 (H-1′a); 3.95 m, 1 H (H-2′); 3.51 dd, 1 H, *J*(1′b,2′) = 8.8 (H-1′b); 3.13 s, 2 H (CH_2); 3.00 dd, 1 H, *J*(3′a,2′) = 5.1, *J*(gem) = 9.5 (H-3′a); 2.87 dd, 1 H, *J*(3′b,2′) = 5.6 (H-3′b). ^{13}C NMR (DMSO-d_6): 162.97 (C-4); 151.19 (C-2); 142.545 (C-6); 143.955, 3 C (Tr); 139.96 and 128.62, 2 C (Ph); 128.43, 6 C (Tr); 128.34, 2 C (Ph); 128.08, 6 C and 127.22, 3 C (Tr); 126.18 (Ph); 110.48 (C-5); 86.13 (Tr); 67.34 (C-2′); 66.175 (C-3′); 52.72 (C-1′).

Phosphonate Esters **23a**–**23c**. General Procedure

A solution of appropriate hydroxy derivative **22** (2.5 mmol) and **5** (3 mmol) in tetrahydrofuran (30 ml) was cooled to –20 °C and 60% oil dispersion of NaH (150 mg, 3.7 mmol) was added. The mixture was stirred at –20 °C for 15 min, then at room temperature for 7 h and filtered through Celite. The filtrate was evaporated, the residue coevaporated with toluene and chromatographed on a column of silica gel (200 ml) in the system described below.

5-Benzyl-1-{(S)-2-[(diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}-4-methoxypyrimidin-2(1H)-one (**23a**). Chromatography in system chloroform–methanol–triethylamine (100:5:1). Yield 1.12 g (63%), white foam. $[\alpha]_D$ –20.0 (*c* 0.178, $CHCl_3$). For $C_{41}H_{47}N_2O_7P$ (710.8) calculated: 69.28% C, 6.66% H, 3.94% N, 4.36% P; found: 69.29% C, 6.67% H, 3.82% N, 4.40% P. ESI MS, *m/z* (%): 733.3 (100) [M + Na], 243.3 (14) [trityl]. ^{1}H NMR (DMSO-d_6): 7.78 s, 1 H (H-6); 7.39 d, 6 H, 7.33 t, 6 H, 7.26 m, 6 H and 7.18 t, 2 H (H-arom.); 3.98 dd, 1 H, *J*(1′a,2′) = 2.8, *J*(gem) = 11.8 (H-1′a); 3.94 m, 1 H (H-2′); 3.90 dd, 1 H, *J*(1′b,2′) = 7.8 (H-1′b); 3.76 s, 3 H (OCH_3); 3.76 dd, 1 H and 3.68 dd, 1 H, *J*(P,CH) = 9.5,

J(gem) = 13.6 (PCH_2); 3.55 s, 2 H (CH_2); 3.22 dd, 1 H, *J*(3′a,2′) = 2.8, *J*(gem) = 10.6 (H-3′a); 2.96 dd, 1 H, *J*(3′b,2′) = 4.0 (H-3′b). ^{13}C NMR (DMSO-d_6): 169.785 (C-4); 155.44 (C-2); 148.05 (C-6); 143.675, 3 C (Tr); 139.45, 128.48, 2 C and 128.47, 2 C (Ph); 128.43, 6 C, 128.11, 6 C and 127.29, 3 C (Tr); 126.32 (Ph); 105.89 (C-5); 86.37 (Tr); 78.16 d, *J*(P,C) = 12.7 (C-2′); 70.35 d, 2 C, *J*(P,C) = 6.3 (P-O-C); 64.06 d, *J*(P,C) = 165.0 (P-C); 62.95 (C-3′); 54.12 (OCH_3); 50.52 (C-1′); 31.87 (CH_2); 23.95 d and 23.93 d, *J*(P,C) = 3.9 (CH_3); 23.89 d and 23.78 d, *J*(P,C) = 4.9 (CH_3).

1-{(S)-2-[(Diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}-4-methoxy-5-(1-naphthylmethyl)pyrimidin-2(1H)-one (**23b**). Chromatography in system chloroform–methanol–triethylamine (100:2:1). Yield 1.5 g (81%), white foam. $[\alpha]_D$ –17.1 (*c* 0.253, $CHCl_3$). For $C_{45}H_{49}N_2O_7P$ (760.9) calculated: 71.04% C, 6.49% H, 3.68% N, 4.07% P; found: 70.79% C, 6.59% H, 3.47% N, 4.14% P. FAB MS, *m/z* (%): 783.7 (20) [M + Na], 243.3 (100) [trityl]. ^{1}H NMR (DMSO-d_6): 8.07 d, 1 H, 7.94 d, 1 H and 7.80 d, 1 H (H-arom.); 7.66 s, 1 H (H-6); 7.54 m, 2 H, 7.38 t, 1 H, 7.36 d, 6 H, 7.31 t, 6 H, 7.25 t, 3 H and 7.13 d, 1 H (H-arom.); 4.52 d sept, 2 H, *J*(CH,CH_3) = 6.2, *J*(P,OCH) = 7.7 (P-OCH); 4.04 d, 1 H and 4.00 d, 1 H, *J*(gem) = 16.4 (CH_2); 3.95 dd, 1 H, *J*(1′a,2′) = 4.0, *J*(gem) = 12.2 (H-1′a); 3.92 m, 1 H (H-2′); 3.86 dd, 1 H, *J*(1′b,2′) = 6.5 (H-1′b); 3.72 dd, 1 H, *J*(P,CH) = 8.7, *J*(gem) = 13.7 (PCH_a); 3.65 dd, 1 H, *J*(P,CH_b) = 9.2 (PCH_b); 3.17 dd, 1 H, *J*(3′a,2′) = 2.6, *J*(gem) = 10.8 (H-3′a); 2.96 dd, 1 H, *J*(3′b,2′) = 4.4 (H-3′b); 1.18 d, 6 H, 1.14 d, 3 H and 1.13 d, 3 H, *J*(CH_3,CH) = 6.2 (CH_3). ^{13}C NMR (DMSO-d_6): 169.88 (C-4); 155.44 (C-2); 148.36 (C-6); 143.65, 3 C (Tr); 134.90, 133.52, 131.435 and 128.79 (1-naphthyl); 128.41, 6 C, 128.09, 6 C and 127.27, 3 C (Tr); 127.07, 126.43, 125.91, 2 C, 125.76 and 123.58 (1-naphthyl); 104.93 (C-5); 86.38 (Tr); 78.22 d, *J*(P,C) = 11.2 (C-2′); 70.40 d, 2 C, *J*(P,C) = 6.4 (P-O-C); 64.12 d, *J*(P,C) = 161.1 (P-C); 63.05 (C-3′); 50.45 (C-1′); 23.94 d, 23.90 d, 23.84 d and 23.78 d, *J*(P,C) = 3.9 (CH_3).

1-{(S)-2-[(Diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}-4-methoxy-5-(2-naphthylmethyl)pyrimidin-2(1H)-one (**23c**). Chromatography in system chloroform–methanol–triethylamine (100:2:1). Yield 1.1 g (58%), white foam. $[\alpha]_D$ –18.4 (*c* 0.217, $CHCl_3$). For $C_{45}H_{49}N_2O_7P$ (760.9) calculated: 71.04% C, 6.49% H, 3.68% N, 4.07% P; found: 70.51% C, 6.72% H, 3.52% N, 4.27% P. ESI MS, *m/z* (%): 1542.9 (43) [2 M + Na], 783.3 (100) [M + Na], 243.2 (21) [trityl]. ^{1}H NMR (DMSO-d_6): 7.87–7.79 m, 3 H (H-arom.); 7.83 s, 1 H (H-6); 7.65 m, 1 H, 7.49–7.43 m, 2 H, 7.39 m, 6 H, 7.36 dd, 1 H, 7.32 m, 6 H and 7.25 m, 3 H (H-arom.); 4.53 dh, 2 H, *J*(CH,CH_3) = 6.2, *J*(CH,P) = 7.7 (P-OCH); 4.01 dd, 1 H, *J*(1′a,2′) = 3.3, *J*(gem) = 12.1 (H-1′a); 3.97–3.89 m, 2 H (H-1′b, H-2′); 3.76 s, 3 H (OCH_3); 3.80–3.67 m, 4 H (PCH_2, CH_2-2-naphthyl); 3.22 dd, 1 H, *J*(3′a,2′) = 3.3, *J*(gem) = 10.6 (H-3′a); 2.99 dd, 1 H, *J*(3′b,2′) = 4.3 (H-3′b); 1.18 d, 3 H, 1.17 d, 3 H, 1.15 d, 3 H and 1.13 d, 3 H (CH_3). ^{13}C NMR (DMSO-d_6): 169.84 (C-4); 155.47 (C-2); 148.20 (C-6); 143.66, 3 C (Tr); 137.05, 133.26 and 131.91 (naphthyl); 128.42, 6 C and 128.10, 6 C (Tr); 127.97, 127.65, 2 C and 127.31 (2-naphthyl); 127.28, 3 C (Tr); 126.48, 126.18 and 125.61 (2-naphthyl); 105.72 (C-5); 86.38 (Tr); 78.20 d, *J*(C,P) = 12.4 (C-2′); 70.35 d, 2 C, *J*(C,P) = 6.2 (P-O-C); 64.09 d, *J*(C-P) = 165.3 (P-C); 63.00 (C-3′); 54.14 (OCH_3); 50.64 (C-1′); 32.09 (CH_2); 23.95–23.75 m, 4 C (CH_3).

Cytosine Derivatives **26a**–**26c**. General Procedure

A solution of appropriate 4-methoxy derivative **23a**–**23c** (0.74 mmol) in 30% methanolic ammonia (40 ml) was heated in an autoclave at 110 °C for 14 h. The solution was cooled to room temperature, evaporated and the residue chromatographed on silica gel column (100 ml) in system chloroform–methanol 9:1.

5-Benzyl-1-{(S)-2-[(diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}cytosine (**26a**). Yield 275 mg (53%) of a white foam. $[\alpha]_D$ –26.7 (*c* 0.238, $CHCl_3$). For $C_{40}H_{46}N_3O_6P$ (695.8) calculated: 69.05% C, 6.66% H, 6.04% N, 4.45% P; found: 68.78% C, 6.55% H, 5.86% N, 4.48% P. ESI MS, *m/z* (%): 696.8 (41) [M + H], 243.3 (49) [trityl]. ^{1}H NMR ($CDCl_3$): 7.43 m, 6 H and 7.32–7.15 m, 15 H (H-6, H-arom.); 4.69 m, 2 H (P-OCH); 4.20 dd, 1 H, *J*(1′a,2′) = 4.0, *J*(gem) = 13.6 (H-1′a); 3.92 m, 1 H (H-2′); 3.87 dd, 1 H, *J*(P,CH_a) = 8.9, *J*(gem) = 13.4 (PCH_a); 3.75 dd, 1 H, *J*(1′b,2′) = 7.9 (H-1′b); 3.63 dd, 1 H, *J*(P,CH_b) = 10.0 (PCH_b); 3.62 d, 1 H and 3.54 d, 1 H, *J*(gem) = 16.4 (CH_2); 3.38 dd, 1 H, *J*(3′a,2′) = 3.4, *J*(gem) = 10.6 (H-3′a); 3.10 dd, 1 H, *J*(3′b,2′) = 4.1 (H-3′ b); 1.29 d, 6 H, 1.26 d, 3 H and 1.245 d, 3 H, *J*(CH_3,CH) = 6.2 (CH_3). ^{13}C NMR ($CDCl_3$): 165.29 (C-4); 156.44 (C-2); 146.24 (C-6); 143.53, 3 C (Tr); 136.88 and 129.05, 2 C (Ph); 128.58, 6 C (Tr); 128.12, 2 C (Ph); 127.90, 6 C (Tr); 127.19 (Ph); 127.13, 3 C (Tr); 103.42 (C-5); 86.75 (Tr); 79.25 d, *J*(P,C) = 13.7 (C-2′); 71.00 d, *J*(P,C) = 6.3 and 70.81 d, *J*(P,C) = 6.3 (P-O-C); 65.02 d, *J*(P,C) = 169.4 (PCH_2); 62.59 (C-3′); 51.24 (C-1′); 33.94 (CH_2); 24.09 d and 24.06 d, *J*(P,C) = 3.9 (CH_3); 24.03 d and 23.97 d, *J*(P,C) = 4.9 (CH_3).

1-{(S)-2-[(Diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}-5-(1-naphthylmethyl)cytosine (**26b**). Yield 359 mg (65%) of a white foam. $[\alpha]_D$ –16.4 (*c* 0.520, $CHCl_3$). For $C_{44}H_{48}N_3O_6P$ (745.9) calculated: 70.86% C, 6.49% H, 5.63% N, 4.15% P; found: 70.51% C, 6.56% H, 5.36% N, 4.25% P. ESI MS, *m/z* (%): 1513.1 (100) [2 M + Na], 768.4 (56) [M + Na], 243.2 (16) [trityl]. ^{1}H NMR (DMSO-d_6): 7.95 m, 4 H, 7.81 d, 2 H, 7.55 m, 2 H, 7.38 t, 1 H, 7.36 d, 6 H, 7.30 t, 6 H, 7.25 t, 3 H and 7.08 d, 2 H (H-arom.); 7.06 s, 1 H (H-6); 4.52 m, 2 H (P-OCH); 4.00 d, 1 H and 3.95 d, 1 H, *J*(gem) = 17.1 (CH_2); 3.85 m, 1 H (H-2′); 3.77 dd, 1 H, *J*(1′a,2′) = 5.1, *J*(gem) = 13.6 (H-1′a); 3.69 dd, 1 H, *J*(P,CH) = 9.0, *J*(gem) = 13.8 (PCH_a); 3.65 dd, 1 H, *J*(1′b,2′) = 5.5 (H-1′b); 3.63 dd, 1 H (PCH_b); 3.11 dd, 1 H, *J*(3′a,2′) = 3.4, *J*(gem) = 10.4 (H-3′a); 2.89 dd, 1 H, *J*(3′b,2′) = 4.8 (H-3′b); 1.18 d, 3 H, 1.175 d, 3 H, 1.142 d, 3 H and 1.14 d, 3 H, *J*(CH_3,CH) = 6.2. ^{13}C NMR (DMSO-d_6): 165.39 (C-4); 155.72 (C-2); 145.76 (C-6); 143.67, 3 C (Tr); 134.61, 133.53, 131.64 and 128.68 (1-naphthyl); 128.37, 6 C, 128.03, 6 C and 127.19, 3 C (Tr); 127.03, 126.25, 125.87, 125.73, 125.53 and 123.79 (1-naphthyl); 102.78 (C-5); 86.28 (Tr); 78.65 d, *J*(P,C) = 11.7 (C-2′); 70.30 d, 2 C, *J*(P,C) = 6.3 (P-O-C); 64.23 d, *J*(P,C) = 165.0 (P-C); 63.16 (C-3′); 50.00 (C-1′); 29.60 (CH_2); 23.93 d, *J*(P,C) = 3.9, 23.83 d, *J*(P,C) = 4.4 and 23.78 d, 2 C, *J*(P,C) = 4.9 (CH_3).

1-{(S)-2-[(Diisopropoxyphosphoryl)methoxy]-3-(trityloxy)propyl}-5-(2-naphthylmethyl)cytosine (**26c**). Yield 353 mg (64%) of a white foam. $[\alpha]_D$ –20.5 (*c* 0.235, $CHCl_3$). For $C_{44}H_{48}N_3O_6P$ (745.9) calculated: 70.86% C, 6.49% H, 5.63% N, 4.15% P; found: 69.93% C, 6.54% H, 5.77% N, 3.98% P. ESI MS, *m/z* (%): 1491.2 (71) [2 M + H], 768.3 (100) [M + Na], 243.2 (65) [trityl]. ^{1}H NMR (DMSO-d_6): 7.89–7.78 m, 3 H, 7.69 bs, 1 H, 7.50–7.44 m, 2 H and 7.39 m, 7 H (H-arom.); 7.36 s, 1 H (H-6); 7.31 m, 6 H and 7.25 m, 3 H (H-arom.); 4.54 m, 2 H (P-OCH); 3.93–3.83 m, 2 H (H-1′a, H-2′); 3.79–3.65 m, 5 H (H-1′b, PCH_2, C**H**$_2$-2-naphthyl); 3.17 dd, 1 H, *J*(3′a,2′) = 2.7, *J*(gem) = 10.5 (H-3′a); 2.93 dd, 1 H, *J*(3′b,2′) = 4.5 (H-3′b); 1.19 d, 3 H, 1.18 d, 3 H, 1.16 d, 3 H and 1.15 d, 3 H, *J*(CH_3,CH) = 6.4 (CH_3). ^{13}C NMR (DMSO-d_6): 165.19 (C-4); 155.74 (C-2); 145.80 (C-6); 143.70 (Tr); 136.67, 133.20 and 131.94 (2-naphthyl); 128.41 and 128.07 (Tr); 127.94, 127.65, 127.59 and 127.39 (2-naphthyl); 127.23 (Tr); 126.52, 126.21 and 125.63 (2-naphthyl); 103.71 (C-5); 86.30 (Tr); 78.71 d, *J*(C,P) = 12.5 (C-2′); 70.36 d, 2 C, *J*(C,P) = 6.3 (P-O-C); 64.24 d, *J*(C,P) = 165.5 (P-C); 63.17 (C-3′); 50.18 (C-1′); 32.53 (CH_2); 23.98–23.79 m, 4 C (CH_3).

Phosphonic Acids **24a**–**24c**. General Procedure

A solution of appropriate compound **23** (1.04 mmol) in acetonitrile (10 ml) was set aside with bromotrimethylsilane (1.3 ml, 10 mmol) in the dark at room temperature for 24 h. The solution was evaporated, the residue coevaporated with acetonitrile (10 ml), stirred with a mixture ethanol–water–triethylamine 15:15:1 (31 ml) for 5 min and evaporated. The residue was partitioned between water (50 ml) and ether (50 ml), the aqueous layer was concentrated to ca. 1/3 volume and applied onto a column of DEAE Sephadex A-25 (Cl^- form, activated with 0.02 M triethylammonium hydrogencarbonate; 10 ml). Elution was performed with water (200 ml), followed by linear gradient of triethylammonium hydrogencarbonate (0.02–0.05 M, 500 ml). The UV-absorbing fraction eluted with 0.3 M buffer concentration was evaporated, codistilled with water (3 × 50 ml) and deionized on activated charcoal. Final products were lyophilized as free phosphonic acids or transformed to dilithium salts on Dowex 50 (Li^+ form) and lyophilized. The following compounds were prepared:

5-Benzyl-1-[(S)-3-hydroxy-2-(phosphonomethoxy)propyl]uracil (**24a**). Isolated as dilithium salt. Yield 347 mg (80%), white amorphous solid. $[\alpha]_D$ –3.6 (*c* 0.311, H_2O). For $C_{15}H_{17}Li_2N_2O_7P \cdot 2H_2O$ (418.19) calculated: 43.08% C, 5.06% H, 6.70% N, 7.41% P; found: 43.26% C, 4.84% H, 6.63% N, 7.33% P. HR MS (TOF ES^+): For $C_{15}H_{18}Li_2N_2O_7P$ [M + H] calculated: 383.1172; found: 383.1166. ESI MS, *m/z* (%): 383.4 (100) [M + H]. ^1H NMR (D_2O): 7.56 s, 1 H (H-6); 7.37 m, 2 H and 7.31 m, 3 H (H-arom.); 3.95 dd, 1 H, *J*(1′a,2′) = 4.9, *J*(gem) = 14.3 (H-1′a); 3.87 dd, 1 H, *J*(1′b,2′) = 6.7 (H-1′b); 3.79 dd, 1 H, *J*(3′a,2′) = 3.4, *J*(gem) = 12.2 (H-3′a); 3.74 m, 1 H (H-2′); 3.67 s, 2 H (CH_2); 3.66 dd, 1 H and 3.63 dd, 1 H, *J*(P,CH) = 9.8, *J*(gem) = 12.8 (PCH_2); 3.57 dd, 1 H, *J*(3′b,2′) = 4.6 (H-3′b). ^{13}C NMR (D_2O): 166.065 (C-4); 152.305 (C-2); 144.95 (C-6); 138.97, 128.69, 2 C, 128.42, 2 C and 126.55 (C-arom.); 113.75 (C-5); 79.76 d, *J*(P,C) = 11.2 (C-2′); 66.93 d, *J*(P,C) = 154.8 (P-C); 60.39 (C-3′); 48.51 (C-1′); 31.81 (CH_2).

1-[(S)-3-Hydroxy-2-(phosphonomethoxy)propyl]-5-(1-naphthylmethyl)uracil (**24b**). Isolated as free acid. Yield 188 mg (43%), white amorphous solid. $[\alpha]_D$ was not determined (opalescent solution, H_2O). HR MS (FAB): For $C_{19}H_{22}N_2O_7P$ [M + H] calculated: 421.1165; found: 421.1170. FAB MS, *m/z* (%): 443 (10) [M + Na], 421 (5) [M + H]. ^1H NMR (D_2O): 7.82 d, 1 H, 7.57 d, 1 H, 7.48 d, 1 H, 7.37 t, 1 H, 7.28 t, 2 H and 7.24 d, 1 H (H-arom.); 6.76 s, 1 H (H-6); 3.84 d, 1 H and 3.76 d, 1 H, *J*(gem) = 16.6 (CH_2); 3.75 dd, 1 H, *J*(1′a,2′) = 5.6, *J*(gem) = 13.2 (H-1′a); 3.60 dd, 1 H and 3.55 dd, 1 H, *J*(P,CH) = 9.0, *J*(gem) = 13.6 (PCH_2); 3.50 m, 1 H (H-2′); 3.38 m, 1 H (H-1′b); 3.38 m, 1 H and 3.31 m, 1 H (H-3′). ^{13}C NMR (D_2O): 165.31 (C-4); 151.65 (C-2); 144.21 (C-6); 134.16, 133.48, 131.26, 128.58, 127.22, 126.80, 126.28, 125.82, 125.70 and 123.62 (C-arom.); 112.94 (C-5); 79.64, *J*(P,C) = 8.3 (C-2′); 65.02 d, *J*(P,C) = 161.1 (P-C); 60.14 (C-3′); 48.34 (C-1′); 37.71 (CH_2).

1-[(S)-3-Hydroxy-2-(phosphonomethoxy)propyl]-5-[(2-naphthyl)methyl]uracil (**24c**). Isolated as free acid. Yield 214 mg (49%), yellowish solid. $[\alpha]_D$ –11.4 (*c* 0.129, H_2O). HR MS (FAB): For $C_{19}H_{22}N_2O_7P$ [M + H] calculated: 421.1165; found: 421.1176. FAB MS, *m/z* (%): 421 (20) [M + H]. ^1H NMR (D_2O): 7.78–7.66 m, 3 H, 7.60 bs, 1 H and 7.41–7.31 m, 3 H (H-arom.); 7.29 s, 1 H (H-6); 3.80–3.73 m, 2 H (H-1′a, PCH_a); 3.70 dd, 1 H, *J*(3′a,2′) = 3.5, *J*(gem) = 12.3 (H-3′a); 3.68–3.63 m, 4 H (CH_2, PCH_b, H-2′); 3.61 m, 1 H (H-1′b); 3.50 dd, 1 H, *J*(3′b,2′) = 4.2, *J*(gem) = 12.3 (H-3′b). ^{13}C NMR (D_2O): 166.47 (C-4); 152.69 (C-2); 145.43 (C-6); 137.21, 133.79,132.47, 128.78, 128.16, 128.08, 127.87, 127.24, 126.96 and 126.38 (C-arom.); 114.10 (C-5); 80.52 d, *J*(C,P) = 11.1 (C-2′); 66.03 d, *J*(C,P) = 159.3 (P-C); 60.84 (C-3′); 49.42 (C-1′); 32.60 (CH_2).

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