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Tetrahedron: Asymmetry
Tetrahedron: *Asymmetry* 18 (2007) 2233–2247

Bifunctional acyclic nucleoside phosphonates: synthesis of chiral 9-{3-hydroxy[1,4-bis(phosphonomethoxy)]butan-2-yl} derivatives of purines

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Received 4 August 2007; accepted 14 September 2007

Abstract—We report herein a general method for the synthesis of new types of chiral acyclic nucleoside four-carbon bisphosphonates. The alkylation of 2-amino-6-chloropurine and adenine was performed with (2*S*,3*S*)- or (2*R*,3*R*)-1,4-[bis(diisopropoxyphosphoryl)methoxy]]-3-[(methylsulfonyl)oxy]butan-2-yl benzoate. Alkylations provided (2*R*,3*R*) or (2*S*,3*S*) N^9-substituted nucleobases, which were further converted to other derivatives. These conversions included either a modification of the nucleobase or transformation of the bisphosphonate chain. Subsequent deprotection of the diisopropyl esters with bromotrimethylsilane provided the resulting (2*R*,3*R*)- or (2*S*,3*S*)-bisphosphonic acids.

1. Introduction

Most of the antiviral compounds that are currently used in the treatment of the herpes simplex virus (HSV), human immunodeficiency virus (HIV), hepatitis B virus (HBV), varicella zoster virus (VZV), and cytomegalovirus (CMV) infections can be described as acyclic nucleoside or nucleotide analogues.[1–3] The activity of this class of chemical substances is currently behind ongoing intensive research aimed at cancer therapy and viral diseases treatment.[4,5] It is worth mentioning that the absolute configuration of the compounds studied often play an important role and significantly influences their biological potency.[6]

Among the acyclic nucleoside analogues bearing a phosphonomethyl ether group, 9-(2-phosphonomethoxyethyl)adenine (PMEA, adefovir) was approved as an antiviral agent[7] active both against DNA viruses and retroviruses, including HIV.[8–11]

The discovery of PMEA was the beginning of a search for other similar biologically active compounds. A study of structure–activity relationships (SAR) in the series of newly synthesized phosphonomethoxyalkyl purine and pyrimidine derivatives revealed that several nucleobases substituted by 3-hydroxy-2-(phosphonomethoxy)propyl (HPMP) or a 2-phosphonomethoxypropyl (PMP) moiety [(*R*)-PMPA or (*S*)-HPMPC, respectively; (see Fig. 1)], show a broad spectrum of potent antiviral activity.[12–17]

Recently, attention has turned to the synthesis of a new type of ANPs originating from 2-substituted 4-amino-6-hydroxypyrimidines.[18] In these investigations, a significant activity of 6-[2-(phosphonomethoxy)ethoxy]pyrimidine derivatives, compounds derived from 2,4-diaminopyrimidine[19] and 2-amino-4-hydroxypyrimidine,[19] and their C5-substituted congeners[20,21] was discovered.

Among the products isolated in these studies, bisphosphonates **I** and **II** were also identified (Fig. 2).[18] Despite the fact that these compounds constitute a new class of possible antiviral agents, they have not yet received much attention. The aim of this work was to explore the potential biological activity of such substances, which could also be considered as analogues of nucleotide bisphosphate

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doi:10.1016/j.tetasy.2007.09.021

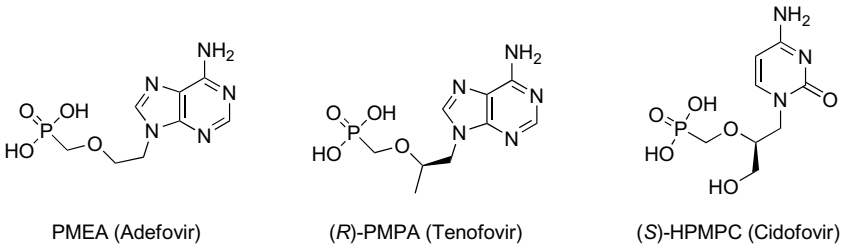


Figure 1.

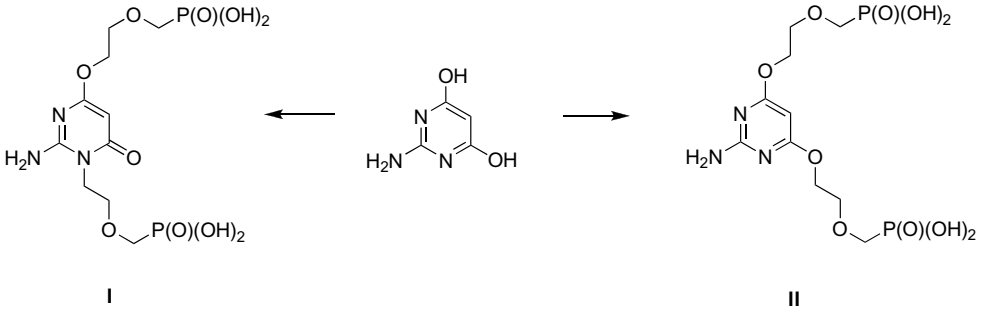


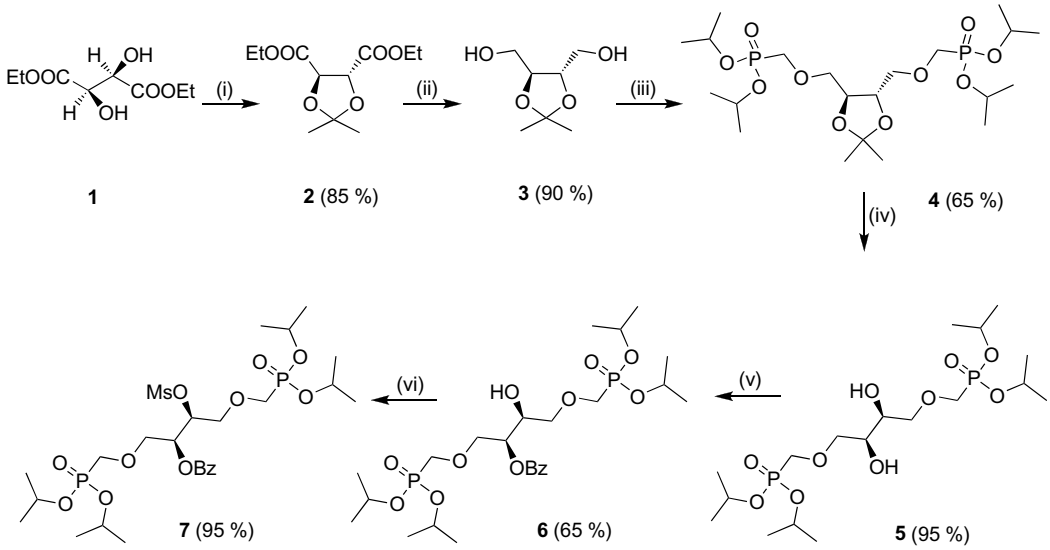
Figure 2.

antagonists of the $P2Y_1$ receptor.[22,23a,b] Additional goal was to study the effect of the introduction of stereogenic centers onto the synthesized compounds, since the chirality seems to play an important role in EI-complex formation in certain enzymes.

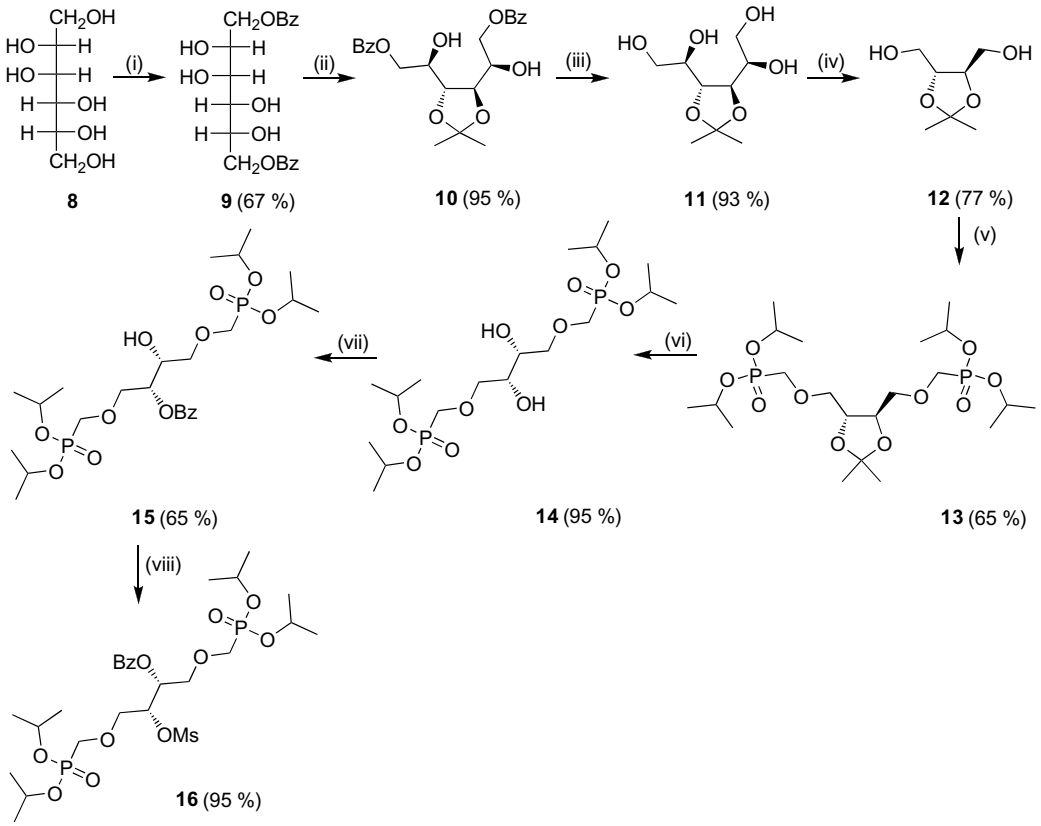
2. Results and discussion

As an outcome of our research, we proposed the following synthetic pathway leading to a novel type of chiral four-carbon acyclic nucleoside bisphosphonates, derivatives of L-threitol and D-mannitol. The strategy of our work was to prepare an appropriate chiral bisphosphonate building block and use it as a common alkylation reagent for various nucleobases. The synthesis of this building block made use of our earlier experience on synthesis of *threo*- and *erythro*-butyl analogues of acyclic nucleosides,[24] it differed for (2*S*,3*S*)- and (2*R*,3*R*)-bisphosphonate enantiomers. While (2*S*,3*S*)-bisphosphonate was prepared from (+)-diethyl L-tartrate **1** (Scheme 1), D-mannitol was used as starting compound for the opposite enantiomer (2*R*,3*R*) (Scheme 2).

The starting (4*S*,5*S*)-(2,2-dimethyl-1,3-dioxolane-4,5-diyl)-dimethanol **3**[24] was prepared from commercially available



Scheme 1. The synthesis of (2*S*,3*S*)-bisphosphonate. Reagents and conditions: (i) $HC(OEt)_3$, 4.5 M HCl/DMF, acetone, rt; (ii) $LiAlH_4$, ether, rt; (iii) NaH, DMF, $TsOCH_2OP(O)(OiPr)_2$, 0 °C, then rt; (iv) Dowex 50 × 8 (H^+ form), 80% isopropyl alcohol; (v) BzCl, pyridine, 0 °C; (vi) MsCl, pyridine, 0 °C.



Scheme 2. The synthesis of (2*R*,3*R*)-bisphosphonate. Reagents and conditions: (i) BzCl, pyridine, 0 °C; (ii) $HC(OEt)_3$, 4.5 M HCl/DMF, acetone, rt; (iii) MeONa/MeOH, MeOH, rt; (iv) $NaIO_4$, $Ba(OAc)_2{\cdot}2H_2O$, then $NaBH_4$, H_2O, acetone; (v) $TsOCH_2P(O)(OiPr)_2$, NaH, DMF, 0 °C; (vi) Dowex 50 × 8 (H^+ form), 80% isopropyl alcohol; (vii) BzCl, pyridine, 0 °C; (viii) MsCl, pyridine, 0 °C.

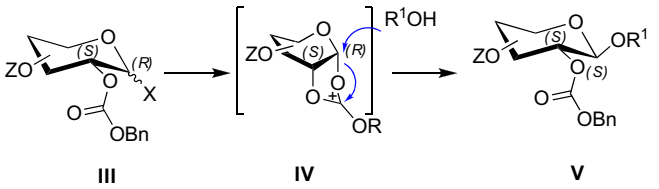
(+)-diethyl L-tartrate **1** as described in Scheme 1. The thus-obtained diol intermediate **3** was alkylated with (diisopropoxyphosphoryl)methyl tosylate[25] and isopropylidene protecting group was subsequently removed with Dowex 50 × 8 (H^+ form) to give compound **5**. The resulting chiral bisphosphonate agent **7** was prepared by mono benzoylation of **5** and subsequent mesylation of **6**. No racemization was observed during this multistep reaction (NMR analysis, specific rotation), so we assumed that the configuration of final chiral four-carbon bisphosphonate was as (2*S*,3*S*).

A multistep synthesis of (2*R*,3*R*)-bisphosphonate was started by the protection of primary hydroxyls of compound **8** (D-mannitol) with benzoyl groups (Scheme 2). *cis*-Oriented hydroxyl groups were then protected by an isopropylidene group. This reaction step was followed by debenzoylation. Cleavage of **11**[26] with $NaIO_4$ in the presence of $Ba(OAc)_2{\cdot}2H_2O$ and $NaBH_4$ reduction gave 3,4-di-*O*-isopropylidene protected 1,4-diol **12**.[26] Alkylating bisphosphonate agent **16** was prepared in the following four steps: attachment of the phosphonate moiety to **12** formed compound **13**, which was then transformed to **14** with Dowex 50 × 8 (H^+ form). Its monobenzoylation provided compound **15**, which gave the final alkylating agent **16** by mesylation. No racemization was observed during this multistep reaction according to NMR analysis, and the specific rotation; therefore, we assigned the configuration of the final chiral four-carbon bisphosphonate as (2*R*,3*R*).

Alkylation of 2-amino-6-chloropurine **17** and adenine **18** with both chiral four-carbon bisphosphonates **7** and **16** provided N^9-substituted derivatives as the only product. However, the mechanism of alkylation and the final configuration at stereocenter C2 could not be unambiguously determined (Scheme 4). We used the benzoyl-protected moiety on stereocenter C3, which is generally used as a participating group for glycosylations in the chemistry of carbohydrates.[27,28] The formation of a single product indicated the possibility of either an S_N2 or S_N1 mechanism. It is known that the participation of the benzoyl group during the glycosylation reaction (S_N1) on the rigid carbohydrate moiety affords the formation of cyclic intermediate **IV**, which allows the nucleophile (e.g., R^1OH) to attack the anomeric carbon only from the opposite side and thus create the highly stereoselective *trans*-product **V** (Scheme 3).

In spite of the fact that free rotation of bisphosphonate chains **7** and **16** probably occurred in our case (Scheme 4), the participation of the benzoyl group could not be excluded.

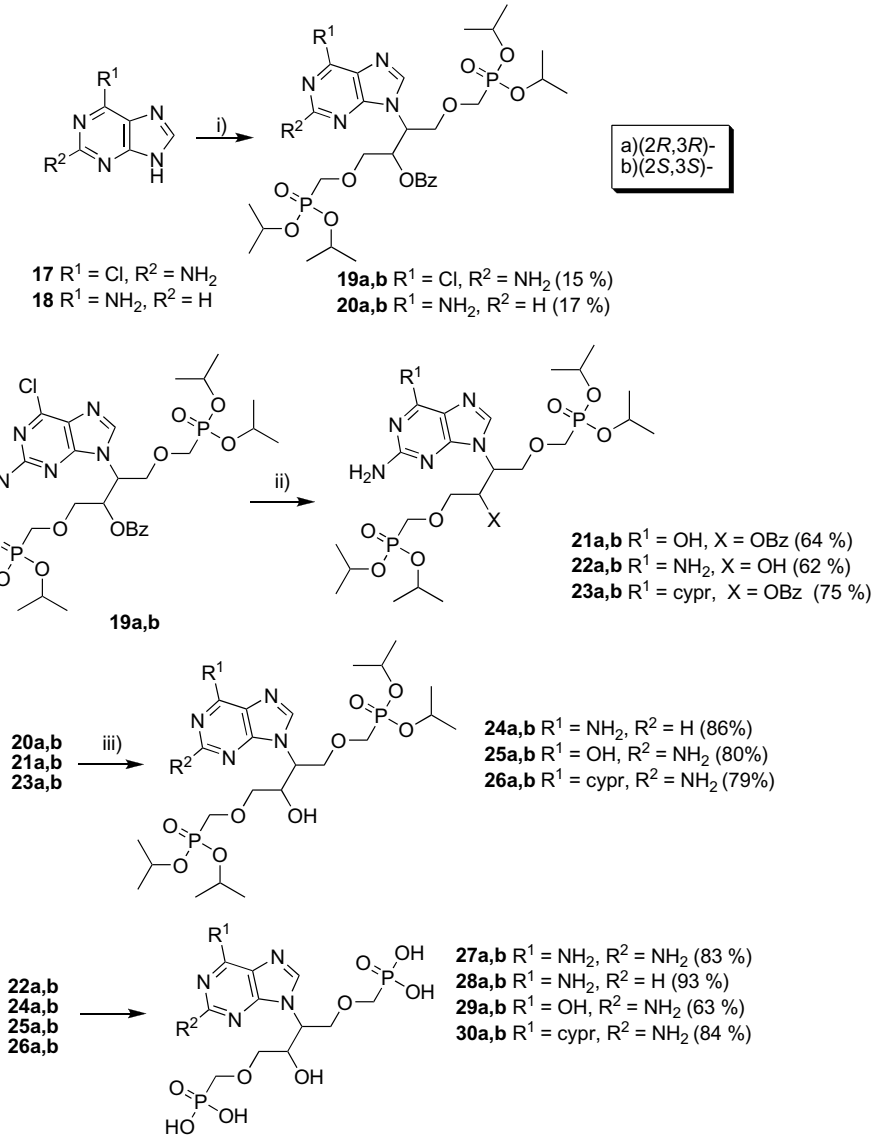
The mechanism of the alkylation reaction was clarified after the replacement of the participating benzoyl group with a non-participating *tert*-butyldimethylsilyl group (Scheme 5). For this purpose compound **33** was prepared and used as the alkylating agent. Alkylation of adenine under the same conditions as used for bisphosphonate **7**



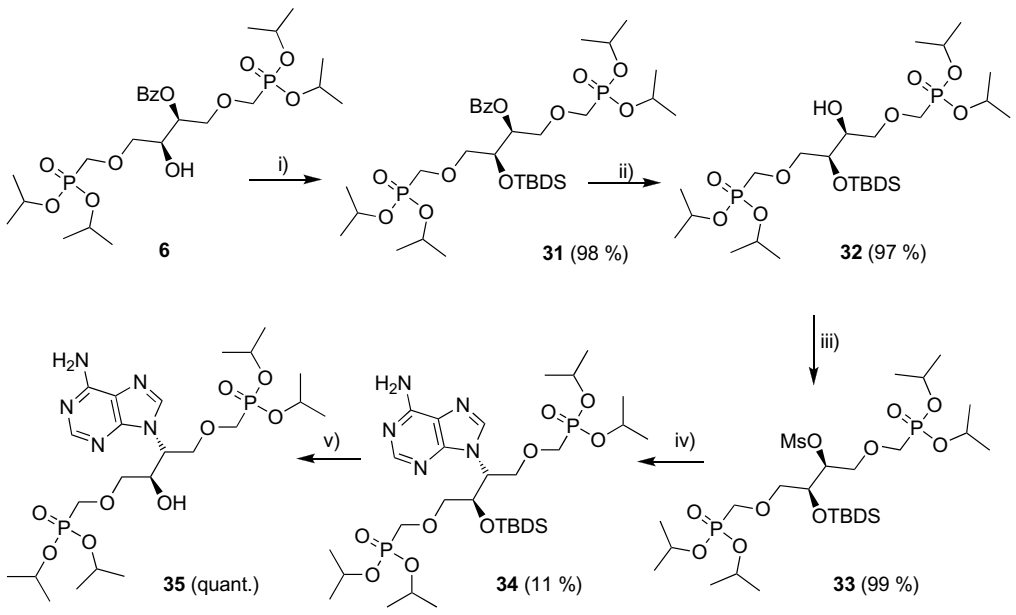
Scheme 3. Glycosylation reaction using participating benzoyl group.

(DMSO, Cs_2CO_3, 110 °C) provided N^9-substituted derivative **34**. Since only one product was isolated, the mechanism was still not certain. Therefore, the protecting silyl group was removed with 1 M TBAF in THF. The structure of the thus-obtained compound **35** was compared with **24a**. An NMR study of both measured derivatives proved that they are identical. Specific rotation analysis also confirmed that **35** was indeed compound **24a**.

NMR spectroscopy, the specific rotation and exclusion of the participation of the silyl group, therefore, revealed that the alkylation of compounds **7**, **16**, and **33** (Schemes 4 and 5) had proceeded by the S_N2 mechanism; that is, the nucleophile (nucleobase) attacks the C2 of bisphosphonates **7**, **16**, and **33** from the opposite side after which the mesyl group leaves and it causes the conversion of configuration on stereocenter C2. As an outcome of this study, this can be said that the alkylation of (2*S*,3*S*)-bisphosphonate **7** gave (2*R*,3*R*)-alkylated derivatives **19a** and **20a**, while opposite enantiomers **19b** and **20b** were formed during the alkylation with (2*R*,3*R*)-bisphoshonate building block **16** (Scheme 4).



Scheme 4. Alkylation of nucleobases with (2*S*,3*S*) or (2*R*,3*R*) bisphosphonate. Reagents and conditions: (i) **7** or **16**, Cs_2CO_3, DMF or DMSO, 110 °C; (ii) for compounds **21a**,**b**: 80% CH_3COOH, reflux; for compounds **22a**,**b**: methanolic ammonia, 100 °C; for compounds **23a**,**b**: cyclopropylamine, dioxane, reflux; (iii) 1 M CH_3ONa/CH_3OH, rt; (iv) TMSBr, CH_3CN, rt.



Scheme 5. The synthesis of (2*S*,3*S*)-bisphosphonate with non-participating group. (i) TBDSCl, imidazole, DMF, rt; (ii) 1 M MeONa/MeOH, rt; (iii) MsCl, pyridine, 0 °C; (iv) adenine **18**, Cs_2CO_3, DMSO, 110 °C; (v) 1 M TBAF/THF, rt.

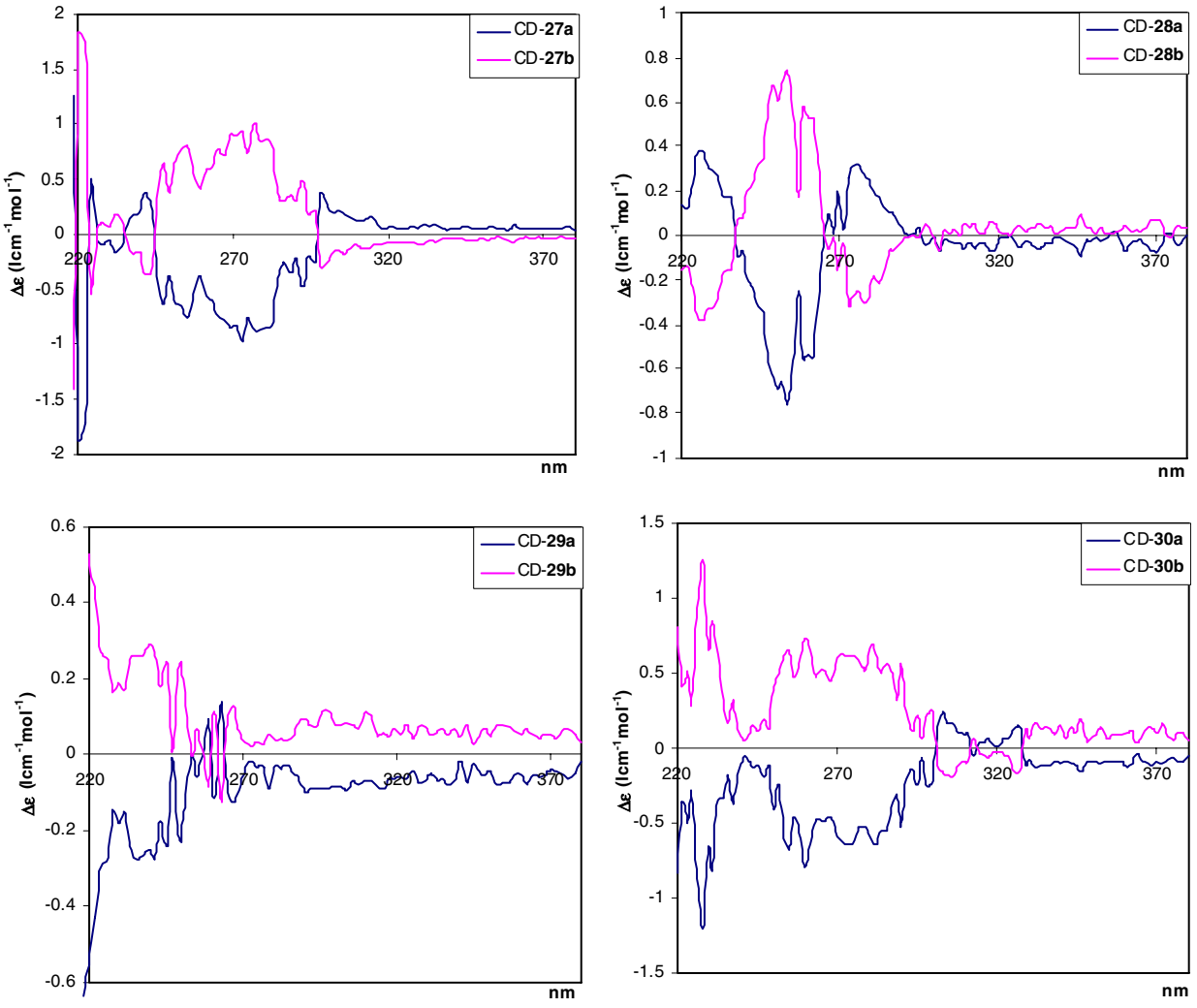


Figure 3. CD spectra of enantiomeric free bisphosphonic acids.

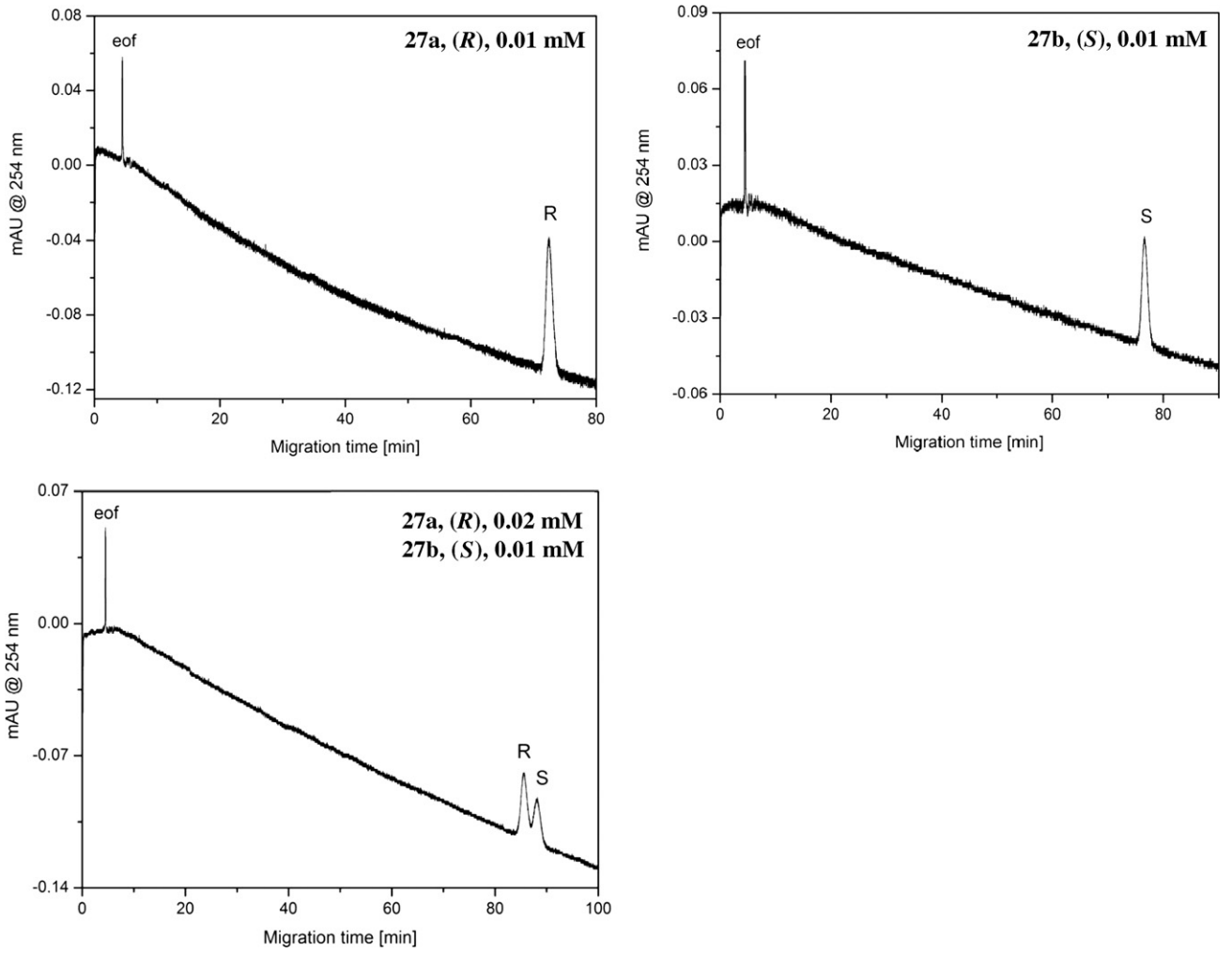


Figure 4. Capillary zone electrophoresis of compounds **27a** and **27b**.

As found by NMR analysis the C3 stereocenter was unchanged during the alkylation (no formation of diastereomer congener was observed).

Transformation of the C–Cl bond of purines **19a**,**b** under standard conditions[23a] afforded the bisphosphonates **21a**,**b**–**23a**,**b** (Scheme 4). Methanolysis catalyzed by sodium methoxide provided compounds **24a**,**b**–**26a**,**b**. Deprotection of the diisopropyl esters with bromotrimethylsilane in acetonitrile, followed by hydrolysis, gave their chiral (2*R*,3*R*)-bisphosphonic acids **27a**–**30a** and (2*S*,3*S*)-bisphosphonic acids **28b** and **29b**, which were isolated from the deionized product by ion exchange chromatography.

Purification of compounds **27b** and **30b** was slightly different. The crude products were applied onto Dowex 50×8 (H^+ form) and washed with water, followed by dilute aqueous ammonia. The thus-obtained ammonium salts were transformed into sodium salts of bisphosphonic acids by Dowex 50×8 (Na^+ form) ion exchange chromatography. Free bisphosphonic acids of original sodium salts of compounds **27b** and **30b** were obtained after the treatment with HCl in water. The enantiomeric purity of final free bisphosphonic acids was among others demonstrated by CD spectra (see Fig. 3) or capillary zone electrophoresis (CZE, Figs. 4 and 5).[29]

3. Conclusion

Chiral four-carbon bisphosphonate alkylating agents were prepared from optically active (+)-diethyl L-tartrate and D-mannitol. These were then used for the synthesis of N^9-alkylated nucleobases, 2-amino-6-chloropurine, and adenine, respectively. As verified in the attempts with non-participating silyl group, the alkylation proceeds via an S_N2 mechanism, which causes an inversion of configuration at the C2 stereocenter. No racemization at the C3 stereocenter took place during the reactions. Therefore, we predicted the configuration of the final bisphosphonic acids to be (2*R*,3*R*) when starting from (2*S*,3*S*)-bisphosphonate, or (2*S*,3*S*) when starting from (2*R*,3*R*)-bisphosphonate.

The enantiomeric purity was confirmed with NMR, CD, or CZE analysis. CZE analyses of compounds **27a**,**b** and **28a**,**b** provided single peaks both in the non-chiral BGE and chiral BGE 2 (see Figs. 4 and 5). No enantioseparation was

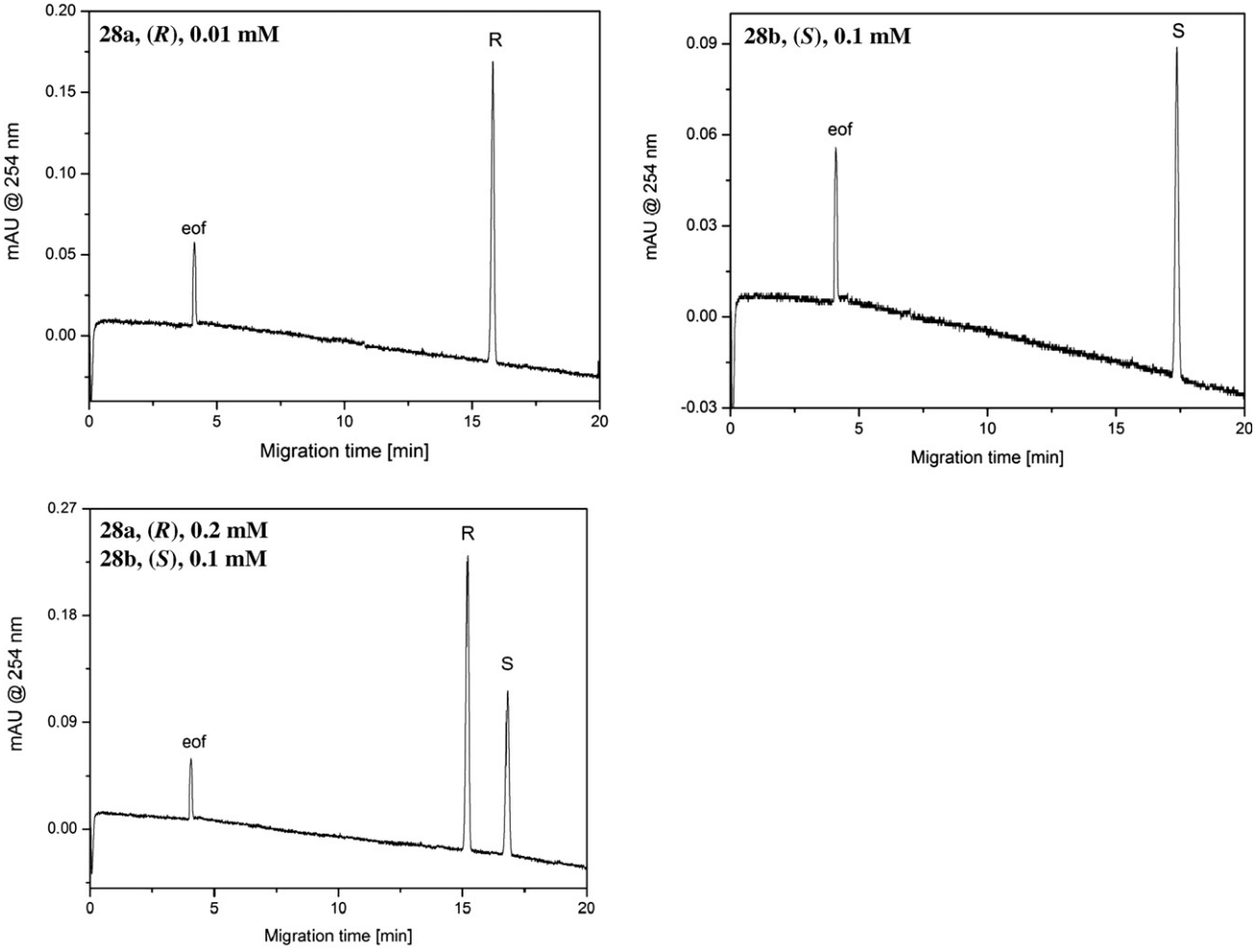


Figure 5. Capillary zone electrophoresis of compounds **28a** and **28b**.

achieved in the case of enantiomeric pairs **29a**,**b** and **30a**,**b**, despite the fact that several chiral BGEs were tested for their separation. This is probably due to the fact that the chiral selector used, β-cyclodextrin, was not able to distinguish among these, because the strength of interactions in both enantiomer–cyclodextrin complexes was the same. When the interaction differs among the enantiomers, it is possible to separate them by means of electrophoresis, as observed for the enantiomeric pairs **27a**,**b** and **28a**,**b**.

The CD spectra and specific rotation indicate that in all cases, the respective enantiomeric pairs were obtained. For all compounds the spectra correspond also quantitatively.

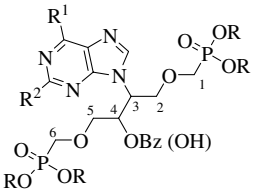
All synthesized compounds are currently undergoing screening for their potential antiviral and cytostatic activity.

4. Experimental

Unless otherwise stated, solvents were evaporated at 40 °C/2 kPa, and compounds were dried over P_2O_5 at 2 kPa. Melting points were determined on a Büchi melting point apparatus and are uncorrected. NMR spectra were measured on a Bruker Avance-500 instrument (500.0 MHz for ^{1}H and 125.7 MHz for ^{13}C). Chemical shifts are given in ppm (δ-scale), and coupling constants (J) in Hz. Mass spectra were measured on a ZAB-EQ spectrometer (Micromass, Manchester, UK) using FAB (ionization by Xe, accelerating voltage 8 kV) or using EI (electron energy 70 eV). UV spectra (λ in nm) were taken on a Beckman Coulter™, DU® 800 spectrophotometer. Elemental analyses were carried out on a Perkin–Elmer 2400 Series II CHNS/O Analyser. Optical rotations were measured on Autopol IV (Rudolph Research Analytical). CD spectra were recorded on Jasco J-810 spectrometer in silicon cells with thickness 0.5–1.0 mm. Aqueous solution with approximate concentration of 0.002 mol/L was used. The results were averaged over three scans (point spacing 0.5 nm, time constant 2 s) due to unfavorable CD signal/absorption ratio. Capillary zone electrophoresis analyses were performed in a commercial P/ACE MDQ apparatus (Beckman Coulter™, Fullerton, CA, USA), equipped with an internally non-coated fused silica capillary with outer polyimide coating (Polymicro Technologies, Phoenix, AR, USA). The analyses were performed both in non-chiral and chiral background electrolytes (BGEs). Chemicals were purchased from Sigma–Aldrich (Prague, Czech Republic).

Dimethylformamide and acetonitrile were distilled from P_2O_5 and stored over molecular sieves (4 Å). Acetone was dried over anhydrous $CuSO_4$. Diethylether was distilled from $LiAlH_4$. Pyridine was dried over KOH and distilled with $KMnO_4$. Methanol was distilled over magnesium pellets.

Numbering for NMR analysis



4.1. (4*R*,5*R*)-Diethyl 2,2-dimethyl-1,3-dioxolane-4,5-dicarboxylate 2

A mixture of (+)-diethyl L-tartrate (309 g; 1.5 mol), dry acetone (450 mL), ethyl orthoformate (332 mL; 1.3 equiv), and 4.5 M HCl in DMF (9 mL) was set aside at room temperature under a $CaCl_2$ cap for 3 days. After neutralization with Et_3N, the mixture was evaporated. The residue was diluted with diethylether (900 mL), washed with water (2×300 mL), dried over $MgSO_4$, filtrated, and evaporated. Distillation in vacuo afforded 310 g (85%) of pure **2** as a yellowish oil, bp 84–86 °C/0.1 Torr. $[\alpha]_D^{20} = +41.2$ (*c* 1, MeOH). FABMS: 247.2 (MH^+) (55). ^{1}H NMR (500 MHz, DMSO-d_6): 4.81 (s, 2H, O–CH); 4.18 (q, 4H, $J_{CH_2,CH_3} = 7.1$, O–CH_2); 1.38 (s, 6H, CH_3); 1.21 (t, 6H, $J_{CH_3,CH_2} = 7.1$, CH_3). ^{13}C NMR (125.7 MHz, DMSO-d_6): 169.60 (2C, CO); 113.04 (C-*i*Pr); 76.74 (2C, O–CH); 61.46 (2C, O–CH_2); 26.48 and 14.08 (2C and 2C, CH_3-*i*Pr and CH_3).

4.2. (4*S*,5*S*)-(2,2-Dimethyl-1,3-dioxolane-4,5-diyl)dimethanol 3

(4*R*,5*R*)-Diethyl 2,2-dimethyl-1,3-dioxolane-4,5-dicarboxylate **2** (269 g, 1.1 mol) in dry diethylether (300 mL) was added dropwise to a stirred and ice-cooled solution of Red-Al (564 mL, 65 wt % solution in toluene) in absolute diethylether (500 mL). The temperature was held below 35 °C. The mixture was then stirred at room temperature for 3 h. The excess hydride was decomposed with ethyl acetate, ethanol, water, and 4 M NaOH. The solids were filtered off and the solution evaporated. The residue was diluted with hot water and neutralized with HCl. The solids were again filtered through the Celite pad and the filtrate washed twice with diethylether. The water layer was evaporated and co-evaporated with ethanol. The residue was diluted with ethyl acetate and the solids filtered through the Celite pad. The organic layer was dried over $MgSO_4$, filtered, and evaporated. The residue was distilled, yielding 108 g (90%) of **3** as a yellowish oil; bp 91–93 °C/0.1 Torr. $[\alpha]_D^{20} = +10.8$ (*c* 0.5, MeOH); FABMS: 163.0 (MH^+) (50). ^{1}H NMR (500 MHz, DMSO-d_6): 4.65 (br s, 2H, OH); 3.74 (m, 2H, O–CH); 3.51 (dd, 2H, $J = 4.1$ and 11.6, O–CH_2); 3.47 (dd, 2H, $J = 5.2$ and 11.6, O–CH_2); 1.30 (s, 6H, CH_3). ^{13}C NMR (125.7 MHz, DMSO-d_6): 108.20 (C-*i*Pr); 78.99 (2C, O–CH); 62.14 (2C, O–CH_2); 27.31 (2C, CH_3).

4.3. (4*S*,5*S*)-[4,5-Bis(diisopropoxyphosphoryl)methoxymethyl]-2,2-dimethyl-1,3-dioxolane 4

A solution of (4*S*,5*S*)-(2,2-dimethyl-1,3-dioxolane-4,5-diyl)dimethanol **3** (20 g, 124 mmol) in dry DMF (200 mL) was added dropwise to a stirred suspension of NaH (12.4 g of 60% suspension in mineral oil, prewashed with *n*-hexane, 310 mmol) in dry DMF (300 mL) at 0 °C under a $CaCl_2$ protecting tube. (Diisopropoxyphosphoryl)methyl tosylate (91 g, 260 mmol) was added dropwise and the mixture stirred at room temperature overnight. The reaction mixture was neutralized with 4.5 M HCl in DMF and the solvent evaporated. The residue was co-evaporated with toluene, dissolved in ethyl acetate (300 mL), and washed with water (3×300 mL). The organic layer was dried over anhydrous $MgSO_4$, filtered, and evaporated. The residue was purified by silica gel column chromatography, using chloroform–methanol gradient 0–2%, to yield 42 g (65%) of pure **4** as a yellowish oil. $[\alpha]_D^{20} = +39.4$ (*c* 0.43, MeOH); FABMS: 519.0 (MH^+) (100). ^{1}H NMR (500 MHz, DMSO-d_6): 4.60 (m, 4H, CH-*i*Pr); 3.91 (m, 2H, O–CH); 3.78 (dd, 4H, $J_{P,CH} = 8.3$, P–CH_2); 3.67 (dd, 2H, $J = 3.1$ and 10.7, O–CH_2); 3.62 (dd, 2H, $J = 5.1$ and 10.7, O–CH_2); 1.31 (s, 6H, CH_3); 1.25 and 1.24 ($4 \times$ d, 24H, $J_{CH_3,CH} = 6.2$, CH_3). ^{13}C NMR (125.7 MHz, DMSO-d_6): 108.99 (C-*i*Pr); 76.50 (2C, O–CH); 72.67 (d, 2C, $J_{P,C} = 11.7$, O–CH_2); 70.31 (d, 4C, $J_{P,C} = 6.4$, CH-*i*Pr); 65.48 (d, 2C, $J_{P,C} = 164.5$, P–CH_2); 27.01 (2C, CH_3); 23.99 (d, 4C, $J_{P,C} = 3.9$, CH_3); 23.88 (d, 4C, $J_{P,C} = 4.4$, CH_3).

4.4. (2*S*,3*S*)-[1,4-Bis(diisopropoxyphosphoryl)methoxy]-butane-2,3-diol 5

A solution of (4*S*,5*S*)-[4,5-bis(diisopropoxyphosphoryl)-methoxymethyl]-2,2-dimethyl-1,3-dioxolane **4** (42 g, 81 mmol) in 80% isopropanol (300 mL) was refluxed with Dowex 50×8 in H^+ form (5 g) for 12 h. The resin was filtered off, the solution neutralized with aqueous ammonia and the solvent evaporated. The crude product was purified by silica gel column chromatography, using chloroform–methanol gradient 0–5%, to yield 37.5 g (95%) of pure **5** as a colorless oil. $[\alpha]_D^{20} = +2.2$ (*c* 0.35, MeOH); FABMS: 479.0 (MH^+) (100). ^{1}H NMR (500 MHz, DMSO-d_6): 4.62 (br s, 2H, OH); 4.60 (m, 4H, CH-*i*Pr); 3.74 and 3.71 (dd, 4H, $J_{P,C} = 8.2$, $J_{gem} = 13.9$, P–CH_2); 3.57 (m, 4H, OCH_2); 3.43 (m, 2H, OCH); 1.24 and 1.23 ($2 \times$ d, 24H, $J_{CH_3,CH} = 6.2$ and 6.3, CH_3). ^{13}C NMR (125.7 MHz, DMSO-d_6): 74.16 (d, 2C, $J_{P,C} = 11.2$, OCH_2); 70.28 and 70.27 (d, 4C, $J_{P,\ C} = 6.4$, CH-*i*Pr); 69.57 (2C, OCH); 65.38 (d, 2C, $J_{P,C} = 164.5$, PCH_2); 24.03 (d, 4C, $J_{P,C} = 3.9$, CH_3); 23.92 (d, 4C, $J_{P,C} = 4.4$, CH_3).

4.5. (2*S*,3*S*)-3-Hydroxy-1,4-[bis(diisopropoxyphosphoryl)-methoxy]butan-2-yl benzoate 6

Benzoyl chloride (8.7 g, 62 mmol) was added dropwise to a stirred solution of **5** (37.0 g, 77 mmol) in dry pyridine (300 mL) at 0 °C with a $CaCl_2$ protecting tube. The mixture was stirred for 2 h at 0 °C and then at room temperature

overnight. The solvent was evaporated and the residue was purified by column chromatography on silica gel, using chloroform–methanol gradient 0–5%, to yield 29.2 g (65%) of pure **6** as a yellowish oil. $[\alpha]_D^{20} = +5.0$ (*c* 0.1, MeOH); FABMS: 583.1 (MH^+) (100). ^{1}H NMR (500 MHz, DMSO-d_6): 8.05 (d, 2H, $H_{arom.}$); 7.66 (t, 1H, $H_{arom.}$); 7.53 (t, 2H, $H_{arom.}$); 5.26 (br s, 1H, OH); 5.22 (m, 1H, OCH); 4.58 and 4.52 (m, 4H, CH-*i*Pr); 3.88 (m, 1H, OCH); 3.81 (d, 2H, $J = 5.0$, OCH_2); 3.58 (dd, 1H, $J = 4.8$ and 10.2, OCH_2); 3.53 (dd, 1H, $J = 6.2$ and 10.2, OCH_2); 3.77 (dd, 1H, $J_{P,C} = 8.2$, $J_{gem} = 13.9$, P–CH_2); 3.75 (d, 2H, $J_{P,C} = 7.8$, P–CH_2); 3.73 (dd, 1H, $J_{P,C} = 8.2$, $J_{gem} = 13.9$, P–CH_2); 1.23–1.13 (8 × d, 24H, $J_{CH_3,CH} = 6.2$, CH_3). ^{13}C NMR (125.7 MHz, DMSO-d_6): 165.40 (CO); 133.49; 129.99; 129.58 (2C), 128.77 (2C); 73.80 (d, $J_{P,C} = 10.2$, OCH_2); 73.17 (OCH); 71.30 (d, $J_{P,C} = 12.2$, OCH_2); 70.34 and 70.30 (2 × d, $J_{P,C} = 5.9$, CH-*i*Pr); 65.48 and 65.19 (2 × d, $J_{P,C} = 164.1$, P–CH_2); 23.99 (d, 4C, $J_{P,C} = 3.9$, CH_3); 23.80 (d, 4C, $J_{P,C} = 4.4$, CH_3).

4.6. (2*S*,3*S*)-3-(Benzoyloxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl mesylate 7

Mesyl chloride (7.8 g, 68 mmol) was added dropwise to a stirred solution of **6** (28.5 g, 49 mmol) in dry pyridine (200 mL) at 0 °C with a $CaCl_2$ protecting tube. The mixture was stirred for 2 h at 0 °C and then at room temperature overnight. The solvent was evaporated and the residue was purified by column chromatography on silica gel, using chloroform–methanol gradient 0–3%, to yield 29.8 g (95%) of pure **7** as a yellowish oil. $[\alpha]_D^{20} = +27.5$ (*c* 0.2, MeOH); For $C_{26}H_{46}O_{13}P_2S$ (660.65) calcd: C, 47.27; H, 7.02; P, 9.38; S, 4.85. Found: C, 47.23; H, 7.14; P, 9.55; S 4.92. FABMS: 661.20 (M^+) (40). ^{1}H NMR (500 MHz, $CDCl_3$): 8.08 (m, 2H, $H_{arom.}$); 7.59 (m, 1H, $H_{arom.}$); 7.46 (m, 2H, $H_{arom.}$); 5.48 (q, 1H, $J_{CH–Bz,CH–Ms} \sim J_{CH–Bz,CHa} \sim J_{CH–Bz,CHb} = 4.8$, CH–Bz); 5.16 (m, 1H, CH–Ms); 4.72 (m, 4H, CH-*i*Pr); 4.00–3.87 (m, 4H, OCH_2); 3.88–3.68 (m, 4H, P–CH_2); 3.18 (s, 3H, Ms–CH_3); 1.34–1.27 (m, 24H, CH_3); ^{13}C NMR (125.7 MHz, $CDCl_3$): 166.45 (CO); 135.54; 129.95 (2C); 129.15; 128.50 (2C); 79.05 (CH–Ms); 72.09 (d, $J_{C,P} = 11.9$, OCH_2); 71.33–71.13 (m, CH-*i*Pr); 70.78 (d, $J_{C,P} = 10.6$, OCH_2); 70.77 (C–Bz); 66.31 (d, $J_{C,P} = 167.4$ and 168.6, P–CH_2); 38.82 (Ms–CH_3); 24.06–23.93 (m, CH_3).

4.7. (2*S*,3*S*)-1,4-[Bis(diisopropoxyphosphoryl)methoxy]-3-(*tert*-butyldimethylsilyloxy)butan-2-yl benzoate 31

A solution of *tert*-butyldimethylsilyl chloride (5.2 g, 34.4 mmol) in dry DMF (50 mL) was slowly added to a solution of **6** (5.0 g, 8.6 mmol) and imidazole (4.1 g, 60.0 mmol) in dry DMF (150 mL). The reaction mixture was stirred overnight at room temperature. It was then concentrated under reduced pressure, dissolved in chloroform, and washed with 0.1 M aq HCl (twice). The organic layer was dried over $MgSO_4$, filtered, and concentrated under reduced pressure. Column chromatography (silica gel, chloroform–methanol gradient 1–5%) afforded product **31** as a yellowish oil (5.87 g, yield 98%). $[\alpha]_D^{20} = +5.0$ (*c* 0.25, MeOH); FABMS: 697.7 (MH^+) (90). ^{1}H NMR (500 MHz, $CDCl_3$): 8.04 (m, 2H, Bz-2); 7.56 (m, 1H, Bz-4); 7.44 (m, 2H, Bz-3); 5.34 (m, 1H, H-3); 4.70 (m, 4H, CH-*i*Pr); 4.13 (m, 1H, H-4); 3.89 (m, 2H, H-2); 3.84–3.69 (m, 4H, H-1 and H-6); 3.65 (m, 2H, H-5); 1.34–1.23 (m, 24H, CH_3-*i*Pr); 0.88 (s, 9H, C–$(CH_3)_3$); 0.11 and 0.09 (2 × s, 6H, CH_3–Si). ^{13}C NMR (125.7 MHz, $CDCl_3$): 165.69 (CO); 133.06 (Bz-4); 130.02 (Bz-1); 129.74 (Bz-2); 128.32 (Bz-3); 74.61 (d, $J_{5,P} = 10.6$, C-5); 73.43 (C-3); 71.25 (d, $J_{2,P} = 11.8$, C-2); 71.03 (m, 4 × CH-*i*Pr); 70.33 (C-4); 66.49 and 66.04 (2 × d, $J_{C,P} = 167.2$ and 167.6, C-1 and C-6); 25.74 (C–$(CH_3)_3$); 24.00 (m, 8 × CH_3-*i*Pr); 17.98 (*C*–$(CH_3)_3$); −4.48 (CH_3–Si).

4.8. (2*S*,3*S*)-1,4-[Bis(diisopropoxyphosphoryl)methoxy]-3-(*tert*-butyldimethylsilyloxy)-butane-2-ol 32

See general procedure for debenzoylation. Column chromatography (silica gel, chloroform–methanol gradient 1–5%) afforded product **32** as a yellowish oil (4.4 g, yield 97%). $[\alpha]_D^{20} = +8.5$ (*c* 0.50, MeOH); FABMS: 593.7 (MH^+) (90). ^{1}H NMR (500 MHz, $CDCl_3$): 4.75 (m, 4H, CH-*i*Pr); 3.90 (m, 1H, H-4); 3.84 (m, 1H, H-3); 3.75 (m, 4H, H-1 and H-6); 3.67 (dd, 1H, $J_{gem} = 9.5$, $J_{5,4} = 6.0$, H-5); 3.64 (dd, 1H, $J_{gem} = 9.9$, $J_{2,3} = 5.3$, H-2); 3.59 (dd, 1H, $J_{gem} = 9.9$, $J_{2',3} = 6.9$, H-2′); 3.57 (dd, 1H, $J_{gem} = 9.5$, $J_{5',4} = 5.9$, H-5′); 1.34 (m, 24H, CH_3-*i*Pr); 0.89 (s, 9H, C–$(CH_3)_3$); 0.10 and 0.09 (2 × s, 6H, CH_3–Si). ^{13}C NMR (125.7 MHz, $CDCl_3$): 74.39 and 74.37(2 × d, $J_{C,P} = 10.6$ and 10.4, C-5 and C-2); 71.02 (m, 4 × CH-*i*Pr); 70.60 (C-4); 70.24 (C-3); 66.23 and 66.22 (2 × d, $J_{C,P} = 167.4$ and 167.7, C-1 and C-6); 25.81 (C–$(CH_3)_3$); 24.00 (m, 8 × CH_3-*i*Pr); 18.05 (*C*–$(CH_3)_3$); −4.38 and −5.03 (CH_3–Si).

4.9. (2*S*,3*S*)-1,4-[Bis(diisopropoxyphosphoryl)methoxy]-3-(*tert*-butyldimethylsilyloxy)butan-2-yl mesylate 33

See mesylation of compound **6**. Column chromatography (silica gel, chloroform–methanol gradient 0–2%) afforded product **33** as a yellowish oil (4.7 g, yield 99%). $[\alpha]_D^{20} = +25.0$ (*c* 0.43, MeOH); FABMS: 671.7 (MH^+) (90). For $C_{25}H_{56}O_{12}P_2SSi$ (670.8) calcd: C, 44.76; H, 8.41; P, 9.23; S, 4.78; Si, 4.19. Found: C, 44.68; H, 8.39; P, 9.19; S, 4.86. ^{1}H NMR (500 MHz, $CDCl_3$): 4.74 (m, 5H, CH-*i*Pr and H-3); 4.03 (m, 1H, H-4); 3.84 (m, 2H, H-2); 3.79 and 3.71 (2 × m, 4H, H-1 and H-6); 3.65 (m, 1H, H-5); 3.14 (s, 3H, Ms–CH_3); 1.33 (m, 24H, CH_3-*i*Pr); 0.89 (s, 9H, C–$(CH_3)_3$); 0.11 (s, 6H, CH_3–Si). ^{13}C NMR (125.7 MHz, $CDCl_3$): 81.42 (C-3); 73.27 and 71.91 (2 × d, $J_{C,P} = 10.8$ and 13.3, C-5 and C-2); 71.06 (m, 4 × CH-*i*Pr); 70.77 (C-4); 66.36 and 66.22 (2 × d, $J_{C,P} = 167.5$ and 169.3, C-1 and C-6); 38.54 (Ms–CH_3); 25.73 (C–$(CH_3)_3$); 24.04 (m, 8 × CH_3-*i*Pr); 17.98 (*C*–$(CH_3)_3$); −5.08 and −4.60 (CH_3–Si).

4.10. 1,6-Di-*O*-benzoyl-D-mannitol 9

Benzoyl chloride (308.6 g, 2196 mmol) was added dropwise to a stirred solution of D-mannitol **8** (200 g, 1098 mmol) in dry pyridine (850 mL) at 0 °C with a $CaCl_2$ protecting tube. The mixture was stirred at 0 °C for 2 h and then at room temperature overnight. The solvent was evaporated

and co-evaporated with toluene. The residue was slowly poured into vigorously stirred water. The white precipitate was filtered and dried in air. Crystallization from ethanol yielded 288 g (67%) of **9** as a white powder. Mp 180–181 °C (lit. 182 °C); $[\alpha]_D^{20} = +15.4$ (*c* 0.5, acetone). For $C_{20}H_{22}O_8$ (390.38) calcd: C, 61.53; H, 5.68. Found: C, 61.63; H, 5.67. FABMS: 391.12 (MH^+) (100). ^{1}H NMR (500 MHz, DMSO-d_6): 7.95 (d, 4H, $H_{arom.}$); 7.56 (t, 2H, $H_{arom.}$); 7.40 (t, 4H, $H_{arom.}$); 4.47 and 4.45 (2 × dd, 2H, $J = 1.4$ and 11.4, H_b-1,6); 4.30 and 4.27 (2 × dd, 2H, $J = 5.8$ and 11.4, H_a-1,6); 3.82 and 3.80 (2 × m, 2H, H-2,5); 3.75 and 3.73 (2 × d, 2H, $J = 9.4$, H-3,4); ^{13}C NMR (125.7 MHz, DMSO-d_6): 165.92 (CO); 132.89, 130.10, 129.15, 128.75, 70.56 (2C, CH-3,4); 68.54 (2C, CH_2); 67.15 (CH-2,5).

4.11. 1,6-Dibenzoyl-3,4-di-*O*-isopropylidene-D-mannitol 10

A mixture of **9** (287 g; 735 mmol), dry acetone (600 mL), ethyl orthoformate (159 mL; 1.3 equiv) and 4.5 M HCl in DMF (5 mL) was set aside for 7 days at room temperature under a $CaCl_2$ protecting tube. After neutralization with Et_3N, the mixture was evaporated, the residue diluted with ethyl acetate (900 mL) and washed with water (2 × 300 mL), dried over $MgSO_4$, filtrated, and evaporated. Crystallization from ethylacetate–petrolether afforded 242 g (95%) of **10** as a white powder. Mp 95–96 °C. $[\alpha]_D^{20} = +32.3$ (*c* 1, MeOH); For $C_{23}H_{26}O_8$ (430.45) calcd: C, 64.18; H, 6.09. Found: C, 63.92; H, 6.05. FABMS: 431.2 (MH^+) (100). ^{1}H NMR (500 MHz, DMSO-d_6): 8.02 (d, 4H, $H_{arom.}$); 7.65 (t, 2H, $H_{arom.}$); 7.52 (t, 4H, $H_{arom.}$); 5.59 (d, 1H, $J_{OH,CH} = 5.4$, OH); 4.47 and 4.45 (2 × dd, 2H, $J = 3.2$ and 11.4, H_b-1,6); 4.30 and 4.26 (2 × dd, 2H, $J = 6.7$ and 11.4, H_a-1,6); 4.10 and 4.05 (2 × d, 2H, $J = 9.4$, H-3,4); 3.94 and 3.92 (2 × m, 2H, H-2,5); 1.34 (s, 3H, CH_3); ^{13}C NMR (125.7 MHz, DMSO-d_6): 165.97 (CO); 133.49, 130.01, 129.48 (2C); 128.87 (2C); 109.44 (C-*i*Pr); 79.54 and 69.82 (2C, CH–OH); 66.59 (O–CH_2); 27.55 (CH_3).

4.12. 3,4-Di-*O*-isopropylidene-D-mannitol 11

A catalytic amount of sodium methoxide (1 M) in methanol (10 mL) was added to a solution of **10** (241 g; 560 mmol) in dry methanol (500) and stirred at room temperature for 5 h. After neutralization with Dowex 50 × 8 (H^+ form), the mixture was filtered, and Dowex was washed with hot methanol. The solution was evaporated and the residue was diluted with water and washed with diethylether. The water layer was evaporated and co-evaporated with ethanol to give 115.6 g (93%) of **11** as a white powder. Mp 86–87 °C (lit. 85–88 °C). $[\alpha]_D^{20} = +31.2$ (*c* 0.8, MeOH). For $C_9H_{18}O_6$ (222.24) calcd: C, 48.64; H, 8.16. Found: C, 48.58; H, 8.27. FABMS: 223.12 (MH^+) (100). ^{1}H NMR (500 MHz, DMSO-d_6): 5.08 (d, 2H, $J_{OH,CH} = 4.6$, OH); 4.46 (t, 2H, $J_{OH,CH_2} = 5.7$, OH); 3.85 and 3.47 (2 × dd, 2 × 2H, O–CH-3,4); 3.54 (ddd, 2H, $J = 3.1$ and 5.6 and 11.2, O–CH_2); 3.35 (dt, 2H, $J = 6.0$ and 6.0 and 11.2, O–CH-2,5); 1.28 (s, 6H, CH_3); ^{13}C NMR (125.7 MHz, DMSO-d_6): 108.48 (C-*i*Pr); 79.25 and 73.07 (4C, OCH); 63.18 (2C, OCH_2); 27.44 (CH_3).

4.13. (4*R*,5*R*)-(2,2-Dimethyl-1,3-dioxolane-4,5-diyl)dimethanol 12

$NaIO_4$ (225 g) was dissolved in hot water (830 mL) and then cooled to 40 °C. Acetone (1000 mL) was added and the reaction mixture was cooled to 15 °C. 3,4-Di-*O*-isopropylidene-D-mannitol **11** (115 g, 517 mmol) was added and the reaction mixture stirred at 10–15 °C for 2 h. The suspension was filtered off and washed with acetone. Acetone was evaporated from the water–acetone solution and $Ba(OAc)_2{\cdot}2H_2O$ (11 g dissolved in minimum water) was added. The precipitate was filtered through the Celite pad and the residue was cooled to 5 °C. $NaBH_4$ (24 g) was added portionwise while keeping the temperature under 20 °C. The reaction mixture was stirred at 15 °C for 2 h and at room temperature overnight. The solution was neutralized with acetic acid, evaporated to 150 mL of final volume, and continually extracted with chloroform for 15 h to yield 65 g (77%) of **12** as a colorless oil. The analysis data correspond with compound **3**. $[\alpha]_D^{20} = -11.2$ (*c* 0.52, MeOH).

4.14. (4*R*,5*R*)-[4,5-Bis(diisopropoxyphosphoryl)methoxymethyl]-2,2-dimethyl-1,3-dioxolane 13

The synthetic procedure and analysis data correspond with compound **4**. $[\alpha]_D^{20} = -38.9$ (*c* 0.50, MeOH).

4.15. (2*R*,3*R*)-[1,4-Bis(diisopropoxyphosphoryl)methoxy]-butane-2,3-diol 14

The synthetic procedure and analysis data correspond with compound **5**. $[\alpha]_D^{20} = -2.4$ (*c* 0.25, MeOH).

4.16. (2*R*,3*R*)-3-Hydroxy-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl benzoate 15

The synthetic procedure and analysis data correspond with compound **6**. $[\alpha]_D^{20} = -5.2$ (*c* 0.30, MeOH).

4.17. (2*R*,3*R*)-3-(Benzoyloxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl mesylate 16

The synthetic procedure and analysis data correspond with compound **7**. $[\alpha]_D^{20} = -27.8$ (*c* 0.45, MeOH).

5. General procedure for alkylation of nucleobases

A solution of an appropriate nucleobase (**17**, **18**, 2 equiv) in dry DMF or DMSO (50 mL) was treated with Cs_2CO_3 (1 equiv) at room temperature under a $CaCl_2$ protecting tube for 1 h. The reaction mixture was then heated at 60 °C and bisphosphonate **7**, **16**, or **33** (0.5 equiv) was added portionvise. The mixture was stirred at 110 °C for 48 h. The solvent was evaporated and the residue co-evaporated with toluene (twice). The residue in 10% MeOH in chloroform was filtered through a Celite pad, evaporated, and purified on a silica gel column in chloroform–methanol.

5.1. (2*R*,3*R*)-2-Amino-6-chloro-9-{3-(benzoyloxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}purine 19a

Column chromatography (silica gel, chloroform–methanol gradient 1–3%) afforded product **19a** as a yellowish oil (2 g, yield 15%). $[\alpha]_D^{20} = +21.8$ (*c* 0.3, MeOH); FABMS: 735.21 (MH^+) (100). ^1H NMR (500 MHz, $CDCl_3$): 8.05–8.02 (m, 3H, Pu-8 and Bz-2); 7.64–7.59 (m, 1H, Bz-4); 7.49–7.45 (m, 2H, Bz-3); 5.75 (dt 1H, $J_{4,5} \sim J_{4,5'} = 3.7$, $J_{4,3} = 8.2$, H-4); 5.30 (br s, 2H, NH_2); 5.23 (dt, 1H, $J_{3,4} \sim J_{3,2} = 7.7$, $J_{3,2'} = 3.4$; H-3); 4.78-4.57 (m, 4H, CH-*i*Pr); 4.40 (dd, 1H, $J_{gem} = 10.2$, $J_{2,3} = 7.4$, H-2); 4.02 (dd, 1H, $J_{gem} = 10.2$, $J_{2',3} = 3.4$, H-2′); 3.84 (dd, 1H, $J_{gem} = 11.1$, $J_{5,4} = 3.6$, H-5); 3.70–3.59 (m, 4H, H-1 and H-6); 3.41 (dd, 1H, $J_{gem} = 11.1$, $J_{5',4} = 3.8$, H-5′); 1.30–1.17 (m, 24 H, CH_3-*i*Pr). ^{13}C NMR (125.7 MHz, $CDCl_3$): 165.19 (CO); 159.10 (Pu-2); 153.90 (Pu-4); 151.40 (Pu-6); 142.63 (Pu-8); 133.63 (Bz-4); 129.81 (Bz-2); 129.25 (Bz-1); 128.59 (Bz-3); 125.02 (Pu-5); 71.42–71.18 (m, 4 × CH-*i*Pr); 70.60 (C-4); 70.59 (d, $J_{5,P} = 12.2$, C-5); 70.26 (d, $J_{2,P} = 9.8$, C-2); 66.38 and 66.24 (2 × d, $J_{C,P} = 169.0$ and 167.5, C-1 and C-6); 54.68 (C-3); 24.03–23.90 (m, 8 × CH_3-*i*Pr).

5.2. (2*S*,3*S*)-2-Amino-6-chloro-9-{3-(benzoyloxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}purine 19b

The synthetic procedure and analysis data correspond with compound **19a**. $[\alpha]_D^{20} = -21.2$ (*c* 0.4, MeOH).

5.3. (2*R*,3*R*)-9-{3-(Benzoyloxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}adenine 20a

Column chromatography (silica gel, chloroform–methanol gradient 1–6%) afforded product **20a** as a yellowish oil (0.42 g, yield 17%). $[\alpha]_D^{20} = +18.7$ (*c* 0.32, MeOH); FABMS: 700.45 (MH^+) (90). ^1H NMR (500 MHz, DMSO-d_6): 8.17 (s, 1H, Pu-8); 8.09 (s, 1H, Pu-2); 7.97 (m, 2H, Bz-2); 7.69 (m, 1H, Bz-4); 7.54 (m, 2H, Bz-3); 7.25 (br s, 2H, NH_2); 5.77 (dt 1H, $J_{4,5} \sim J_{4,5'} = 3.3$, $J_{4,3} = 6.4$, H-4); 5.14 (dt, 1H, $J_{3,4} \sim J_{3,2} = 7.3$, $J_{3,2'} = 3.3$; H-3); 4.55 (m, 4H, CH-*i*Pr); 4.37 (dd, 1H, $J_{gem} = 10.2$, $J_{2,3} = 6.8$, H-2); 4.06 (dd, 1H, $J_{gem} = 10.5$, $J_{2',3} = 4.0$, H-2′); 3.83 (dd, 1H, $J_{gem} = 11.0$, $J_{5,4} = 4.0$, H-5); 3.72 (dd, 1H, $J_{gem} = 11.0$, $J_{5',4} = 4.1$, H-5); 3.70 and 3.67 (2 × d, 2 × 2H, $J_{H,P} = 8.7$ and 8.2, H-1 and H-6); 1.15–1.04 (m, 24H, CH_3-*i*Pr). ^{13}C NMR (125.7 MHz, DMSO-d_6): 164.89 (CO); 156.17 (Pu-6); 152.53 (Pu-2); 149.77 (Pu-4); 140.25 (Pu-8); 133.76 (Bz-4); 129.60 (Bz-2); 129.29 (Bz-1); 128.80 (Bz-3); 118.91 (Pu-5); 71.01 (C-4); 70.72 (d, $J_{5,P} = 10.7$, C-5); 70.37–70.27 (m, 4 × CH-*i*Pr); 69.46 (d, $J_{2,P} = 11.7$, C-2); 65.25 and 64.99 (2 × d, $J_{C,P} = 164.1$ and 163.6, C-1 and C-6); 54.38 (C-3); 23.90–23.58 (m, 8 × CH_3-*i*Pr).

5.4. (2*S*,3*S*)-9-{3-(Benzoyloxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}adenine 20b

The synthetic procedure and analysis data correspond with compound **20a**. $[\alpha]_D^{20} = -18.8$ (*c* 0.4, MeOH).

5.5. (2*R*,3*R*)-9-{1,4-[Bis(diisopropoxyphosphoryl)methoxy]butan-2-yl-3-(*tert*-butyldimethylsilyloxy)}adenine 34

Column chromatography (silica gel, chloroform–methanol gradient 2–3%) afforded product **34** as a yellowish oil (0.31 g, yield 11%). $[\alpha]_D^{20} = +18.2$ (*c* 0.59, MeOH); FABMS: 700.2 (MH^+) (90). ^1H NMR (500 MHz, $CDCl_3$): 8.31 (s, 1H, Pu-2); 8.04 (s, 1H, Pu-8); 5.70 (br s, 2H, NH_2); 4.87 (ddd 1H, $J_{3,2'} = 3.7$, $J_{3,2} \sim J_{3,4} = 6.8$ and 8.2, H-3); 4.72–4.60 and 4.61–4.54 (2 × m, 4H, CH-*i*Pr); 4.38 (m, 2H, H-2 and H-4); 3.97 (dd, 1H, $J_{gem} = 10.1$, $J_{2',3} = 3.7$, H-2′); 3.69–3.54 (m, 4H, H-1 and H-6); 3.47 (dd, 1H, $J_{gem} = 10.3$, $J_{5,4} = 4.6$, H-5); 3.28 (dd, 1H, $J_{gem} = 10.3$, $J_{5',4} = 4.8$, H-5′); 1.29–1.12 (m, 24H, CH_3-*i*Pr); 0.90 (s, 9H, C–$(CH_3)_3$); 0.07 and −0.04 (2 × s, 6H, CH_3–Si). ^{13}C NMR (125.7 MHz, $CDCl_3$): 155.26 (Pu-6); 152.64 (Pu-2); 150.05 (Pu-4); 141.35 (Pu-8); 119.39 (Pu-5); 74.34 (d, $J_{5,P} = 9.7$, C-5); 71.00 (m, 4 × CH-*i*Pr); 70.25 (C-4); 70.00 (d, $J_{2,P} = 11.5$, C-2); 66.31 and 65.98 (2 × d, $J_{C,P} = 167.1$ and 168.1, C-1 and C-6); 56.91 (C-3); 25.77 (C-$(CH_3)_3$); 23.90 (m, 8 × CH_3-*i*Pr); −4.36 and −5.28 (CH_3–Si).

5.6. (2*R*,3*R*)-9-{3-(Benzoyloxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}guanine 21a

A solution of **19a** (0.6 g, 0.8 mmol) in 80% acetic acid (25 mL) was refluxed for 6 h. The solution was neutralized with Et_3N and the volatiles were evaporated in vacuo. Column chromatography (silica gel, chloroform–methanol gradient 1–12%) afforded compound **21a** as a yellowish oil (0.37 g, yield 64%). $[\alpha]_D^{20} = +20.3$ (*c* 0.3, MeOH); FABMS: 716.32 (MH^+) (50). ^1H NMR (500 MHz, $CDCl_3$): 11.78 (br s, 1H, NH); 8.10–8.01 (m, 2H, Bz-2); 7.78 (s, 1H, Pu-8); 7.64–7.56 (m, 1H, Bz-4); 7.50–7.43 (m, 2H, Bz-3); 6.54 (br s, 2H, NH_2); 5.72 (dt 1H, $J_{4,5} \sim J_{4,5'} = 3.8$, $J_{4,3} = 8.2$, H-4); 5.09 (dt, 1H, $J_{3,4} \sim J_{3,2} = 7.3$, $J_{3,2'} = 3.3$; H-3); 4.77-4.59 (m, 4H, CH-*i*Pr); 4.33 (dd, 1H, $J_{gem} = 10.2$, $J_{2,3} = 6.8$, H-2); 4.02 (dd, 1H, $J_{gem} = 10.2$, $J_{2',3} = 3.3$, H-2); 3.84 (dd, 1H, $J_{gem} = 11.0$, $J_{5,4} = 4.0$, H-5); 3.53 (dd, 1H, $J_{gem} = 11.0$, $J_{5,4} = 4.1$, H-5); 3.71 and 3.69 (2 × d, 2 × 2H, $J_{H,P} = 8.7$ and 8.2, H-1 and H-6); 1.29–1.19 (m, 24H, 8 × CH_3-*i*Pr). ^{13}C NMR (125.7 MHz, $CDCl_3$): 165.18 (CO); 158.91 (Pu-6); 153.74 (Pu-2); 151.70 (Pu-4); 137.34 (Pu-8); 133.56 (Bz-4); 129.82 (Bz-2); 129.35 (Bz-1); 128.57 (Bz-3); 116.92 (Pu-5); 71.55–71.27 (m, 4 × CH-*i*Pr); 70.88 (d, $J_{2,P} = 9.7$, C-2); 70.85 (C-4); 70.78 (d, $J_{5,P} = 12.6$, C-5); 66.38 and 66.24 (2 × d, $J_{C,P} = 169.4$ and 167.5, C-1 and C-6); 53.94 (C-3); 23.87, 23.88–24.05 (m, 8 × CH_3-*i*Pr).

5.7. (2*S*,3*S*)-9-{3-(Benzoyloxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}guanine 21b

The synthetic procedure and analysis data correspond with compound **19a**. $[\alpha]_D^{20} = -20.2$ (*c* 0.35, MeOH).

5.8. (2*R*,3*R*)-2,6-Diamino-9-{3-(hydroxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}purine 22a

A solution of **19a** (0.6 g, 0.8 mmol) in methanolic ammonia (50 mL) was heated (100 °C) in an autoclave for 13 h. The solvent was then evaporated. Purification of the residue by

column chromatography (silica gel, chloroform–methanol gradient 1–11%) afforded compound **22a** as a yellowish foam (0.30, yield 62%). $[\alpha]_D^{20} = +17.3$ (*c* 0.23, MeOH + 2 drops $CHCl_3$); FABMS: 611.58 (MH^+) (90). ^{1}H NMR (500 MHz, $CDCl_3$): 7.63 (s, 1H, Pu-8); 6.04 (br s, 1H, OH); 5.62 (br s, 2H, 6-NH_2); 4.81 (br s, 2H, 2-NH_2); 4.80–4.62 (m, 4H, CH-*i*Pr); 4.59 (dt, 1H, $J_{3,4} \sim J_{3,2} = 7.5$, $J_{3,2'} = 3.4$; H-3); 4.32 (dt 1H, $J_{4,5} \sim J_{4,5'} = 3.7$, $J_{4,3} = 8.2$, H-4); 4.13 (dd, 1H, $J_{gem} = 10.3$, $J_{2,3} = 7.7$, H-2); 3.99 (dd, 1H, $J_{gem} = 10.2$, $J_{2',3} = 3.3$, H-2′); 3.83 and 3.77 (2 × d, 2 × 2H, $J_{H,P} = 8.7$ and 8.2, H-1 and H-6); 3.72 (dd, 1H, $J_{gem} = 11.0$, $J_{5,4} = 4.0$, H-5); 3.65 (dd, 1H, $J_{gem} = 11.0$, $J_{5',4} = 4.1$, H-5′); 1.34–1.24 (m, 24H, CH_3-*i*Pr). ^{13}C NMR (125.7 MHz, $CDCl_3$): 159.08 (Pu-2); 156.00 (Pu-6); 151.08 (Pu-4); 139.04 (Pu-8); 114.38 (Pu-5); 74.16 (d, $J_{5,P} = 10.0$, C-5); 71.24–71.09 (m, 4 × CH-*i*Pr); 70.83 (d, $J_{2,P} = 10.5$, C-2); 70.20 (C-4); 66.41 and 66.05 (2 × d, $J_{C,\ P} = 165.0$ and 163.6, C-1 and C-6); 57.68 (C-3); 24.08–23.87 (m, 8 × CH_3-*i*Pr).

5.9. (2*S*,3*S*)-2,6-Diamino-9-{3-(hydroxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}purine 22b

The synthetic procedure and analysis data correspond with compound **22a**. $[\alpha]_D^{20} = -17.5$ (*c* 0.20, MeOH).

5.10. (2*R*,3*R*)-2-Amino-9-{3-(benzoyloxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}-6-(cyclopropyl)aminopurine 23a

A solution of **19a** (0.5 g, 0.68 mmol) in dry dioxane (20 mL) was refluxed with cyclopropylamine (0.38 mL, 5.44 mmol) for 13 h. The solvent was then evaporated. Purification of the residue by column chromatography (silica gel, chloroform–methanol gradient 1–4%) afforded compound **23a** as a yellowish oil (0.38 g, yield 75%). $[\alpha]_D^{20} = +24.5$ (*c* 0.25, $CHCl_3$); FABMS: 755.54 (MH^+) (90). ^{1}H NMR (500 MHz, $CDCl_3$): 8.06 (s, 1H, Bz-2); 7.69 (s, 1H, Pu-8); 7.60 (m, 1H, Bz-4); 7.47 (m, 2H, Bz-3); 5.80 (dt 1H, $J_{4,5} \sim J_{4,5'} = 3.8$, $J_{4,3} = 8.3$, H-4); 5.09 (ddd, 1H, $J_{3,4} = 8.4$, $J_{3,2'} = 7.4$, $J_{3,2} = 3.4$; H-3); 4.83 (br s, 2H, NH_2); 4.74–4.55 (m, 4H, CH-*i*Pr); 4.35 (dd, 1H, $J_{gem} = 10.1$, $J_{2,3} = 7.4$, H-2); 3.97 (dd, 1H, $J_{gem} = 10.1$, $J_{2',3} = 3.4$, H-2′); 3.80 (dd, 1H, $J_{gem} = 11.0$, $J_{5,4} = 3.3$, H-5); 3.70 and 3.65 (2 × d, 2 × 2H, $J_{H,P} = 8.7$ and 8.2, H-1 and H-6); 3.42 (dd, 1H, $J_{gem} = 11.0$, $J_{5',4} = 4.2$, H-5′); 2.98 (br s, 1H, $CH_{cyclopropyl}$); 1.29–1.16 (m, 24H, CH_3-*i*Pr); 0.90–0.82 (m, 2H, CH_2); 0.64–0.59 (m, 2H, CH_2). ^{13}C NMR (125.7 MHz, $CDCl_3$): 165.26 (CO); 159.91 (Pu-2); 156.19 (Pu-6); 151.05 (Pu-4); 137.62 (Pu-8); 133.49 (Bz-4); 129.81 (Bz-2); 129.39 (Bz-1); 128.51 (Bz-3); 114.25 (Pu-5); 71.26–71.07 (m, 4 × CH-*i*Pr); 70.80 (d, $J_{5,P} = 12.3$, C-5); 70.77 (d, $J_{2,P} = 11.3$, C-2); 70.75 (C-4); 66.21 and 66.15 (2 × d, $J_{C,P} = 168.1$ and 167.1, C-1 and C-6); 54.07 (C-3′); 24.02–23.77 (m, 8 × CH_3, $CH_{cyclopropyl}$); 7.32 and 6.47 (2 × CH_2).

5.11. (2*S*,3*S*)-2-Amino-9-{3-(benzoyloxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}-6-(cyclopropyl)aminopurine 23b

The synthetic procedure and analysis data correspond with compound **23a**. $[\alpha]_D^{20} = -24.6$ (*c* 0.20, $CHCl_3$).

6. General procedure for debenzoylation

A catalytic amount of 1 M sodium methoxide in methanol was added to a well-dried benzoylated compound (**20a**,**b**; **21a**,**b**; **23a**,**b**; 0.5–0.6 mmol) in dry methanol and stirred at rt under $CaCl_2$ protecting tube until the full conversion of the starting compound. The mixture was neutralized with HCl or CH_3COOH, evaporated, and purified on a silica gel column in chloroform–methanol.

6.1. (2*R*,3*R*)-9-{3-(Hydroxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}adenine 24a

Column chromatography (silica gel, chloroform–methanol gradient 1–8%) afforded product **24a** as a yellowish oil (0.31 g, yield 86%). $[\alpha]_D^{20} = +12.4$ (*c* 0.60, MeOH); FABMS: 596.35 (MH^+) (80). ^{1}H NMR (500 MHz, DMSO-d_6): 8.12 (s, 1H, Pu-8); 8.10 (s, 1H, Pu-2); 7.20 (br s, 2H, NH_2); 5.63 (d, 1H, $J_{OH,4} = 5.8$, OH); 4.72 (ddd, 1H, $J_{3,2'} = 3.8$, $J_{3,4} = 6.6$, $J_{3,2} = 9.2$, H-3); 4.58 and 4.38 (2 × m, 4H, CH-*i*Pr); 4.25 (t, 1H, $J_{2,3} = J_{2,2'} = 9.8$, H-2a); 4.13 (m, 1H, H-4); 4.01 (dd, 1H, $J_{2',2} = 10.5$, $J_{2',\ 3} = 3.7$, H-2′); 3.80–3.64 (2 × d, 2 × 2H, $J_{H,P} = 8.8$ and 8.4, H-1 and H-6); 3.45 (dd, 1H, $J_{gem} = 11.2$, $J_{5,4} = 4.0$, H-5); 3.36 (dd, 1H, $J_{gem} = 11.2$, $J_{5',4} = 4.1$, H-5′); 1.23–1.02 (m, 24 H, CH_3-*i*Pr). ^{13}C NMR (125.7 MHz, DMSO-d_6): 156.17 (Pu-6); 152.32 (Pu-2); 149.68 (Pu-4); 140.38 (Pu-8); 118.91 (Pu-5) 74.21 (d, $J_{5,P} = 10.0$, C-5); 70.34 (m, 4 × CH-*i*Pr); 70.22 (d, $J_{2,P} = 12.2$, C-2); 68.86 (C-4); 65.44 (d, $J_{6,P} = 163.5$, C-6); 64.94 (d, $J_{1,P} = 163.6$, C-1); 55.93 (C-3); 24.02–23.67 (m, 8 × CH_3-*i*Pr).

6.2. (2*S*,3*S*)-9-{3-(Hydroxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}adenine 24b

The synthetic procedure and analysis data correspond with compound **24a**. $[\alpha]_D^{20} = -12.6$ (*c* 0.60, MeOH).

6.3. (2*R*,3*R*)-9-{3-(Hydroxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}guanine 25a

Column chromatography (silica gel, chloroform–methanol gradient 1–8%) afforded product **25a** as a yellowish oil (0.26 g, yield 80%). $[\alpha]_D^{20} = +18.2$ (*c* 0.2, $CHCl_3$); FABMS: 612.23 (MH^+) (50). ^{1}H NMR (500 MHz, DMSO-d_6): 10.73 (br s, 1H, NH); 7.68 (s, 1H, Pu-8); 6.59 (br s, 2H, NH_2); 5.60 (d, 1H, $J_{OH,H\text{-}4} = 5.8$, OH); 4.62–4.52 (m, 4H, CH-*i*Pr); 4.40 (dt, 1H, $J_{3,4} \sim J_{3,2} = 7.5$, $J_{3,2'} = 3.4$; H-3); 4.12 (dd, 1H, $J_{gem} = 10.4$, $J_{2,3} = 6.6$, H-2); 4.03 (dt 1H, $J_{4,5} \sim J_{4,5'} = 3.7$, $J_{4,3} = 8.2$, H-4); 3.94 (dd, 1H, $J_{gem} = 10.5$, $J_{2',3} = 3.6$, H-2′); 3.77 and 3.61 (2 × d, 2 × 2H, $J_{H,P} = 8.7$ and 8.2, H-1 and H-6); 3.45 (dd, 1H, $J_{gem} = 11.0$, $J_{5,4} = 4.0$, H-5); 3.40 (dd, 1H, $J_{gem} = 11.0$, $J_{5',4} = 4.1$, H-5); 1.23–1.10 (m, 24H, CH_3-*i*Pr). ^{13}C NMR (125.7 MHz, DMSO-d_6): 156.83 (Pu-6); 153.63 (Pu-2); 151.16 (Pu-4); 136.72 (Pu-8); 116.54 (Pu-5); 74.13 (d, $J_{C,P} = 12.6$, C-5); 70.50–70.20 (m, 4 × CH-*i*Pr, C-2); 68.86 (C-4); 65.48 and 65.01 (2 × d, $J_{C,P} = 165.0$ and 163.6, C-1 and C-6); 55.52 (C-3); 24.00–23.60 (m, 8 × CH_3-*i*Pr).

6.4. (2*S*,3*S*)-9-{3-(Hydroxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}guanine 25b

The synthetic procedure and analysis data correspond with compound **25a**. $[\alpha]_D^{20} = -18.9$ (*c* 0.20, $CHCl_3$).

6.5. (2*R*,3*R*)-2-Amino-6-(cyclopropyl)amino-9-{3-(hydroxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}purine 26a

Column chromatography (silica gel, chloroform–methanol gradient 1–5%) afforded product **26a** as a yellowish oil (0.26 g, yield 79%). $[\alpha]_D^{20} = +19.9$ (*c* 0.22, $CHCl_3$); FABMS: 651.45 (MH^+) (80). ^1H NMR (500 MHz, $CDCl_3$): 7.63 (s, 1H, Pu-8); 6.07 (br s, 1H, OH); 5.67 (br s, 2H, NH_2); 4.80–4.62 (m, 4H, CH-*i*Pr); 4.60 (ddd, 1H, $J_{3,4} \sim J_{3,2} = 7.5$, $J_{3,2'} = 3.4$, H-3); 4.26 (dt, 1H, $J_{4,5} \sim J_{4,5'} = 3.7$, $J_{4,3} = 8.2$, H-4); 4.15 (dd, 1H, $J_{gem} = 10.1$, $J_{2,3} = 7.5$, H-2); 4.01 (dd, 1H, $J_{gem} = 10.2$, $J_{2',3} = 3.3$, H-2′); 3.83 and 3.77 ($2 \times$ d, 2×2H, $J_{H,P} = 8.7$ and 8.2, H-1 and H-6); 3.72 (dd, 1H, $J_{gem} = 11.0$, $J_{5,4} = 4.0$, H-5); 3.65 (dd, 1H, $J_{gem} = 11.0$, $J_{5',4} = 4.1$, H-5′); 2.95 (br s, 1H, $CH_{cyclopropyl}$); 1.34-1.24 (m, 24H, CH_3-*i*Pr); 0.85 (m, 2H, CH_2); 0.60 (m, 2H, CH_2). ^{13}C NMR (125.7 MHz, $CDCl_3$): 159.72 (Pu-2); 156.35 (Pu-6); 151.18 (Pu-4); 139.24 (Pu-8); 114.48 (Pu-5) 74.14 (d, $J_{5,P} = 10.0$, C-5); 71.11 (m, $4 \times$ CH-*i*Pr); 70.81 (d, $J_{2,P} = 10.5$, C-2); 70.20 (C-4); 66.41 (d, $J_{6,P} = 163.5$, C-6); 66.05 (d, $J_{1,P} = 163.6$, C-1); 57.68 (C-3); 24.05–23.87 (m, $8 \times CH_3$, $CH_{cyclopropyl}$); 7.31 and 6.49 ($2 \times CH_2$).

6.6. (2*S*,3*S*)-2-Amino-6-(cyclopropyl)amino-9-{3-(hydroxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}purine 26b

The synthetic procedure and analysis data correspond with compound **26a**. $[\alpha]_D^{20} = -20.1$ (*c* 0.20, $CHCl_3$).

6.7. (2*R*,3*R*)-9-{3-(Hydroxy)-1,4-[bis(diisopropoxyphosphoryl)methoxy]butan-2-yl}adenine 35

An oven-dried round bottomed flask under an argon atmosphere was charged with **34** (110 mg, 0.15 mmol) and 1 M solution of TBAF in THF (0.85 mL, 0.85 mmol). The reaction was aged at rt for 2 h and concentrated. Purification on thin layer chromatography with UV silica gel sorbent (eluent 10% MeOH in $CHCl_3$) afforded pure **35** as a yellowish oil (90 mg, quant. yield). $[\alpha]_D^{20} = +12.7$ (*c* 0.71, MeOH). For $C_{23}H_{43}N_5O_9P_2$ (595.56) calcd: C, 46.38; H, 7.28; N, 11.76; P, 10.40. Found: C, 46.43; H, 7.35; N, 11.72; P, 10.50. Other analysis data (FABMS and NMR) correspond with compound **24a**.

7. Transformation of esters to free phosphonic acids—general procedure

The dried esters (**22a**,**b**;**24a**,**b**;**25a**,**b**;**26a**,**b**; 0.4–0.5 mmol), acetonitrile (15 mL) and $BrSiMe_3$ (2–3 mL) were stirred at room temperature overnight. After evaporation and co-distillation with acetonitrile, the residue was treated with water and aqueous ammonia. The mixture was evaporated to dryness, and the residue dissolved in water applied onto a column of Dowex 50×8 in H^+ form and washed with water.

(A) Elution with water, evaporation in vacuo, and crystallization from water/ethanol afforded free bisphosphonic acids **27a**; **28a**,**b**; **29a**,**b**; **30a**.
(B) Elution with dilute ammonia and evaporation afforded ammonium salts, which were applied onto Dowex 50 (Na^+ form). Elution with water and evaporation gave bisphosphonic acids **27b** and **30b** as tetrasodium salts. Precipitation of free bisphosphonic acids was obtained after the treatment of sodium salts with HCl in water.

7.1. (2*R*,3*R*)-2,6-Diamino-9-{3-(hydroxy)-1,4-[bis(phosphonomethoxy)]butan-2-yl}purine 27a

White solid (0.18 g, yield 83%), mp 142.1 °C (D). $[\alpha]_D^{20} = +19.2$ (*c* 0.34, H_2O). For $C_{11}H_{20}N_6O_9P_2$ (442.26) calcd: C, 29.87; H, 4.56; N, 19.00; P, 14.01. Found: C, 29.95; H, 4.62; N, 19.12; P, 13.95. FABMS: 443.3 (MH^+) (40). ^1H NMR (500 MHz, D_2O): 8.20 (s, 1H, Pu-8); 4.82 (dt, 1H, $J_{3,4} \sim J_{3,2} = 7.4$, $J_{3,2'} = 3.2$, H-3); 4.32 (ddd, 1H, $J_{4,3} = 7.2$, $J_{4,5'} = 5.2$, $J_{4,5} = 3.8$, H-4); 4.30 (dd, 1H, $J_{gem} = 11.0$, $J_{2,3} = 7.7$, H-2); 4.09 (dd, 1H, $J_{gem} = 11.0$, $J_{2',3} = 3.3$, H-2′); 3.80 (dd, 1H, $J_{gem} = 13.3$, $J_{1,P} = 8.8$, H-1); 3.73 (dd, 1H, $J_{gem} = 13.3$, $J_{1',P} = 9.0$, H-1′); 3.65–3.61 (m, 3H, 6, 6′, 5); 3.44 (dd, 1H, $J_{gem} = 10.8$, $J_{5',4} = 5.2$, H-5′). ^{13}C NMR (125.7 MHz, D_2O): 153.03 (Pu-2); 151.02 (Pu-6); 150.94 (Pu-4); 142.27 (Pu-8); 111.17 (Pu-5) 73.74 (d, $J_{5,P} = 12.3$, C-5); 70.53 (d, $J_{2,P} = 12.7$, C-2); 69.22 (C-4); 67.52 and 67.47 ($2 \times$ d, $J_{C,P} = 157.8$ and 157.5, C-1 and C-6); 56.85 (C-3). UV spectrum: (0.01 M HCl) $\lambda_{max} = 250$ nm ($\varepsilon_{max} = 7804$); (H_2O) $\lambda_{max} = 251$ nm ($\varepsilon_{max} = 7286$); (0.01 M NaOH) $\lambda_{max} = 254$ nm ($\varepsilon_{max} = 6562$).

7.2. (2*S*,3*S*)-2,6-Diamino-9-{3-(hydroxy)-1,4-[bis(phosphonomethoxy)]butan-2-yl}purine 27b

The synthetic procedure (followed by method B), NMR and UV data correspond with compound **27a**. White solid (0.12 g, yield 88%). $[\alpha]_D^{20} = -19.0$ (*c* 0.20, H_2O).

7.3. (2*R*,3*R*)-9-{3-(Hydroxy)-1,4-[bis(phosphonomethoxy)]-butan-2-yl}adenine 28a

White solid (0.21 g, yield 93%), mp 147.2 °C (D). $[\alpha]_D^{20} = +12.2$ (*c* 0.26, H_2O). For $C_{11}H_{19}N_5O_9P_2$ (427.24) calcd: C, 30.92; H, 4.48; N, 16.39; P, 14.50. Found: C, 30.98; H, 4.56; N, 16.35; P, 14.61. FABMS: 428.3 (MH^+) (40). ^1H NMR (500 MHz, D_2O): 8.56 (s, 1H, Pu-8); 8.41 (s, 1H, Pu-2); 5.04 (dt, 1H, $J_{3,4} \sim J_{3,2} = 7.5$, $J_{3,2'} = 3.3$, H-3); 4.44 (ddd, 1H, $J_{4,3} = 7.6$, $J_{4,5} = 5.2$, $J_{4,5'} = 3.7$, H-4); 4.34 (dd, 1H, $J_{gem} = 11.1$, $J_{2,3} = 7.4$, H-2); 4.10 (dd, 1H, $J_{gem} = 11.0$, $J_{2',3} = 3.4$, H-2′); 3.73 (dd, 1H, $J_{gem} = 13.4$, $J_{1,P} = 8.8$, H-1); 3.69 (dd, 1H, $J_{gem} = 13.4$, $J_{1',P} = 8.8$, H-1′); 3.63 (m, 1H, H-5); 3.58 (m, 2H, 6 and 6′); 3.45 (dd, 1H, $J_{gem} = 10.8$, $J_{5',4} = 5.2$, H-5′). ^{13}C NMR (125.7 MHz, D_2O): 150.54 (Pu-6); 149.27 (Pu-4); 145.07 (Pu-8); 144.94 (Pu-2); 118.55 (Pu-5) 73.86

(d, $J_{5,P} = 11.7$, C-5); 70.76 (d, $J_{2,P} = 12.2$, C-2); 69.26 (C-4); 67.50 and 67.38 (2 × d, $J_{C,P} = 158.2$ and 157.7, C-1 and C-6); 57.36 (C-3). UV spectrum: (0.01 M HCl) $\lambda_{max} = 257$ nm ($\varepsilon_{max} = 12{,}992$); ($H_2O$) $\lambda_{max} = 258$ nm ($\varepsilon_{max} = 13{,}274$); (0.01 M NaOH) $\lambda_{max} = 259$ nm ($\varepsilon_{max} = 13{,}122$).

7.4. (2*S*,3*S*)-9-{3-(Hydroxy)-1,4-[bis(phosphonomethoxy)]-butan-2-yl}adenine 28b

The synthetic procedure and analysis data correspond with compound **28a**. $[\alpha]_D^{20} = -11.9$ (*c* 0.2, H_2O).

7.5. (2*R*,3*R*)-9-{3-(Hydroxy)-1,4-[bis(phosphonomethoxy)]-butan-2-yl}guanine 29a

White solid (0.12 g, yield 63%), mp 163.3 °C (D). $[\alpha]_D^{20} = +10.9$ (*c* 0.23, H_2O). For $C_{11}H_{19}N_5O_{10}P_2$ (443.24) calcd: C, 29.81; H, 4.32; N, 15.80; P, 13.98. Found: C, 29.92; H, 4.36; N, 15.85; P, 13.87. FABMS: 444.2 (MH^+) (60). ^{1}H NMR (500 MHz, D_2O): 9.08 (br s, 1H, Pu-8); 5.00 (dt, 1H, $J_{3,4} \sim J_{3,2} = 6.9$, $J_{3,2'} = 3.3$, H-3); 4.38 (dt, 1H, $J_{4,3} = 7.1$, $J_{4,5} \sim J_{4,5'} = 4.6$, H-4); 4.29 (dd, 1H, $J_{gem} = 11.0$, $J_{2,3} = 6.6$, H-2); 4.04 (dd, 1H, $J_{gem} = 11.0$, $J_{2',3} = 3.3$, H-2′); 3.74 (m, 2H, H-1); 3.70 (dd, 1H, $J_{gem} = 10.7$, $J_{5,4} = 4.2$, H-5); 3.65 (d, 2H, $J = 6.6$, H-6); 3.58 (dd, 1H, $J_{gem} = 10.7$, $J_{5',4} = 5.0$, H-5′). ^{13}C NMR (125.7 MHz, D_2O): 155.98 and 155.96 (Pu-6 and Pu-2); 150.86 (Pu-4); 138.34 (Pu-8); 108.26 (Pu-5) 73.77 (d, $J_{5,P} = 12.1$, C-5); 69.98 (d, $J_{2,P} = 12.5$, C-2); 68.80 (C-4); 67.43 and 67.39 (2 × d, $J_{C,P} = 158.3$ and 158.0, C-1 and C-6); 57.76 (C-3). UV spectrum: (0.01 M HCl) $\lambda_{max} = 258$ nm ($\varepsilon_{max} = 10{,}960$); ($H_2O$) $\lambda_{max} = 258$ nm ($\varepsilon_{max} = 10{,}968$); (0.01 M NaOH) $\lambda_{max} = 282$ nm ($\varepsilon_{max} = 10{,}796$).

7.6. (2*S*,3*S*)-9-{3-(Hydroxy)-1,4-[bis(phosphonomethoxy)]-butan-2-yl}guanine 29b

The synthetic procedure and analysis data correspond with compound **29a**. $[\alpha]_D^{20} = -10.5$ (*c* 0.16, H_2O).

7.7. (2*R*,3*R*)-2-Amino-6-(cyclopropyl)amino-9-{3-(hydroxy)-1,4-[bis(phosphonomethoxy)]butan-2-yl}purine 30a

White solid (0.16 g, yield 84%), mp 164.3 °C (D). $[\alpha]_D^{20} = +12.9$ (*c* 0.25, H_2O). For $C_{14}H_{24}N_6O_9P_2$ (482.32) calcd: C, 34.86; H, 5.02; N, 17.42; P, 12.84. Found: C, 34.96; H, 4.96; N, 17.35; P, 12.71. FABMS: 483.1 (MH^+) (65). ^{1}H NMR (500 MHz, D_2O): 8.16 (br s, 1H, Pu-8); 4.80 (dt, 1H, $J_{3,4} \sim J_{3,2} = 7.2$, $J_{3,2'} = 3.1$, H-3); 4.34 (ddd, 1H, $J_{4,3} = 7.8$, $J_{4,5} = 3.7$, $J_{4,5'} = 5.1$, H-4); 4.26 (dd, 1H, $J_{gem} = 10.9$, $J_{2,3} = 7.2$, H-2); 4.05 (dd, 1H, $J_{gem} = 10.9$, $J_{2',3} = 3.2$, H-2′); 3.76–3.57 (m, 5H, H-1,1′, H-5, H-6,6′); 3.41 (dd, 1H, $J_{gem} = 10.8$, $J_{5',4} = 5.1$, H-5′); 2.95 (br s, 1H, $CH_{cyclopropyl}$); 1.04 (m, 2H, CH_2); 0.84 (m, 2H, CH_2). ^{13}C NMR (125.7 MHz, D_2O): 153.26 and 151.43 (Pu-2 and Pu-6); 149.62 (Pu-4); 141.95 (Pu-8); 111.71 (Pu-5); 73.74 (d, $J_{5,P} = 12.1$, C-5); 70.72 (d, $J_{2,P} = 12.2$, C-2); 69.20 (C-4); 67.66 and 67.58 (2 × d, $J_{C,P} = 157.7$ and 157.6, C-6 and C-1); 56.72 (C-3); 23.44 (br s, $CH_{cyclopropyl}$); 7.47 (br s, 2C, 2 × CH_2). UV spectrum: (0.01 M HCl) $\lambda_{max} = 253$ nm and 292 nm ($\varepsilon_{max} = 9216$ and 11,408); (H_2O) $\lambda_{max} = 253$ nm and 290 nm ($\varepsilon_{max} = 9156$ and 11,394); (0.01 M NaOH) $\lambda_{max} = 258$ nm and 282 nm ($\varepsilon_{max} = 7590$ and 12,546).

7.8. (2*S*,3*S*)-2-Amino-6-(cyclopropyl)amino-9-{3-(hydroxy)-1,4-[bis(phosphonomethoxy)]butan-2-yl}-purine 30b

The synthetic procedure (followed by method B), NMR and UV data correspond with compound **30a**. White solid (0.11 g, yield 88%). $[\alpha]_D^{20} = -13.1$ (*c* 0.16, H_2O).

Acknowledgments

This work is a part of the research project Z4 055 0506. It was supported by the 'Centre for New Antivirals and Antineoplastics' (1M0508), by the Program of Targeted Projects of Academy of Sciences of the Czech Republic (1QS400550501) and by Gilead Sciences, Inc. (Foster City, CA, USA). We would like to thank the team of Dr. P. Maloň for measurement of CD spectra, and also to Dr. V. Kašička and Dr. V. Šolínová for performing the CZE measurements.

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