ALTERNATIVE SYNTHESIS OF 9-{3-[(DIISOPROPOXYPHOSPHORYL)-METHOXY]-2-HYDROXYPROPYL}ADENINE AND ITS FREE PHOSPHONATES SUBSTITUTED AT THE C-8 POSITION OF PURINE BASE

Zlatko JANEBA[1,*], Milena MASOJÍDKOVÁ[2] and Antonín HOLÝ[3]

Institute of Organic Chemistry and Biochemistry, Academy of Sciences of the Czech Republic, v.v.i., Flemingovo nám. 2, 166 10 Prague 6, Czech Republic; e-mail: [1] janeba@uochb.cas.cz, [2] masojidkova@uochb.cas.cz, [3] holy@uochb.cas.cz

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For its high therapeutic effect, (*S*)-9-[3-hydroxy-2-(phosphonomethoxy)propyl]adenine (HPMPA) is an important member of a class of acyclic nucleoside phosphonates (ANPs). Although its constitutional isomer, 9-[2-hydroxy-3-(phosphonomethoxy)propyl]adenine (iso-HPMPA), exhibits no antiviral activity, our general interest in C-8 substituted adenine ANPs led us to prepare certain iso-HPMPA derivatives modified at the C-8 position of adenine. Novel alkylating agent, diisopropyl {[2-(tetrahydro-2-pyranyl)oxy-3-tosyloxypropoxy]-methyl}phosphonate (**9**), was prepared by procedure starting from allyl alcohol (**4**). 9-{3-[(Diisopropoxyphosphoryl)methoxy]-2-hydroxypropyl}adenine (**12**) was prepared by alkylation of adenine with the alkylating agent **9** followed by acid hydrolysis, although elimination by-product 9-{3-[(diisopropoxyphosphoryl)methoxy]prop-1-enyl}adenine (**11**) predominated in the reaction mixture. Bromination of the compound **12** gave 8-bromo-adenine derivative **13** quantitatively. Nucleophilic substitutions of the bromine atom of compound **13** with *N*- and *O*-nucleophiles, followed by phosphonate deprotection, afforded the free phoshonic acids **15**–**18**.

Keywords: Acyclic nucleoside phosphonates; Acyclic nucleotide analogues; HPMPA; Alkylation; Nucleophilic substitution; Intramolecular cyclization.

Structural modifications of nucleosides and nucleotides have been for a long time significant means of research in medicinal and bioorganic chemistry, biotechnology and other biological applications. Acyclic nucleoside phosphonates (ANPs), or acyclic nucleotide analogues, have certainly occupied very important position in the field[1]. Among other side chain modifications, *N*-[3-hydroxy-2-(phosphonomethoxy)propyl] (HPMP) derivatives[2] of purine and pyrimidine bases have been studied in detail owing to their antiviral properties and were found to be selectively active against DNA

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viruses (herpes viruses, adenoviruses, poxviruses, iridoviruses, polyoma and papilomaviruses)[3]. (*S*)-HPMPC (Cidofovir)[2,4] has been approved for treatment of CMV retinitis in AIDS patients (Vistide™)[5], although it also is efficacious in other viral diseases[6].

While the (*S*)-enantiomer is active in adenine series (HPMPA, **1**, Chart 1)[3], its 3′-*O*-phosphonomethyl isomer (iso-HPMPA, **2**)[7] exhibits no antiviral activity[3a]. Recently, certain ANPs were found to selectively inhibit hypoxanthine-guanine-xanthine phosphoribosyltransferase of *Plasmodium falciparum* and have been studied as potential anti-malarials[8]. These findings revived our interest in the iso-HPMPA analogues, especially those modified at the C-8 position of the purines.

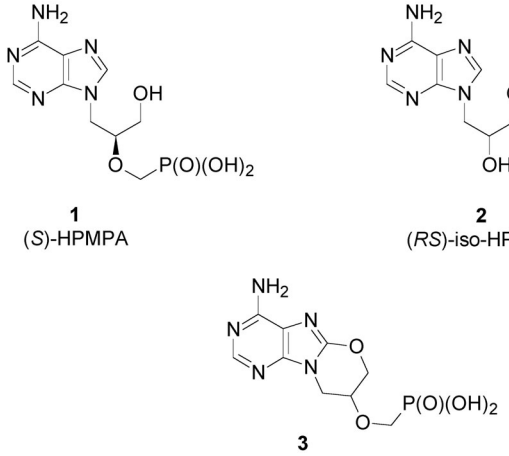


CHART 1

Some 10 years ago, as a part of the Ph.D. Thesis of the first author (Z.J.)[9], a synthesis of racemic 9-{3-[(diisopropoxyphosphoryl)methoxy]-2-hydroxypropyl}adenine was designed starting from allyl alcohol. Later, another synthetic approach to iso-HPMP derivatives of purine bases using oxirane ring opening was developed[10] and immunobiological properties of these compounds were studied[11].

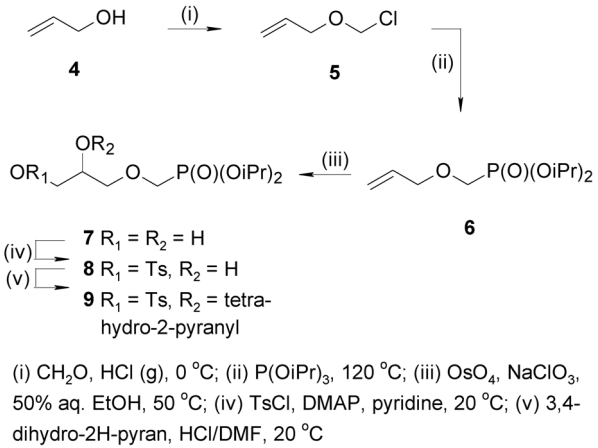
Within the extensive systematic studies of various types of acyclic nucleoside and nucleotide analogues we have paid substantial attention to a group of compounds modified at the C-8 position of purine bases[12]. These compounds showed to be interesting both for their biological properties[13] and for the synthetic and mechanistic studies (e.g. formation and ring opening of 8,*O*-anhydro derivatives, e.g. compound **3**, Chart 1)[12a,12b,14]. The intramolecular cyclizations to form the corresponding anhydro derivatives

could be studied in the case of the HPMP analogues, where the free hydroxyl group in the acyclic moiety can participate in the cyclization process.

Due to possible important biological properties of the modified iso-HPMP analogues we decided to describe the original procedure for the preparation of racemic iso-HPMPA [9]. An additional aim of this study was to prepare 8-substituted iso-HPMPA analogues for SAR studies, namely 8-hydroxy (8-oxo) and *N*-substituted 8-aminoadenine derivatives. Also, compound of special interest was 8,2′-*O*-anhydro analogue, a constitutional isomer of formerly described 8,3′-*O*-anhydro HPMPA derivative **3** (Chart 1)[12b].

RESULTS AND DISCUSSION

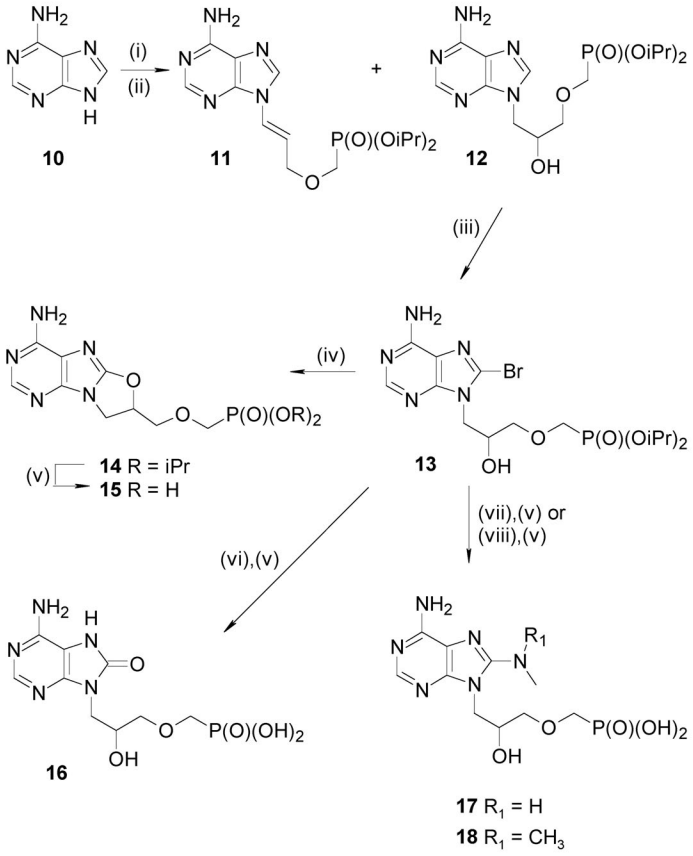
Allyl alcohol (**4**) was chosen as a convenient starting material for the synthesis of suitable alkylating agent leading to iso-HPMPA analogue. Allyl alcohol (**4**) was converted by chloromethylation to chloro derivative **5** [15] which under Arbuzov reaction[16] conditions with triisopropyl phosphite afforded diisopropyl phosphonate **6** [17] in 32% overall yield from the starting alcohol **4** (Scheme 1). The double bond of the compound **6** was hydroxylated under catalysis of OsO_4 to afford 2,3-dihydroxy derivative **7** in moderate yield. The compound **7** was tosylated at the primary hydroxyl to give derivative **8**. Then the secondary hydroxyl of compound **8** was protected by the reaction with 3,4-dihydro-2H-pyran to afford compound **9** in 37% overall yield from **7**.



SCHEME 1

Alkylation of adenine (**10**) with the tosylate **9** using DBU, followed by treatment of the crude alkylation product with 1 M HCl at 60 °C for 2 h

gave a mixture of two products (Scheme 2): the desired product **12** (11%) and compound **11** (31%) which was formed by elimination of the hydroxyl group at C-2′ under the acid conditions. The value of vicinal coupling constants[18] of H-1′ and H-2′ protons (J(1′,2′) = J(2′,1′) = 14.5 Hz) confirmed the *trans* isomer of **11** being the sole product of the elimination process.



(i) **9**, DBU, DMF, 110 °C; (ii) 1M aq. HCl, 60 °C; (iii) Br_2, dioxane, H_2O, $NaHPO_4$, 20 °C; (iv) NaH, DMF, 20 °C; (v) TMSBr, MeCN, 20 °C; (vi) AcONa, AcOH, reflux; (vii) NH_2CH_3, EtOH, 100 °C; (viii) $NH(CH_3)_2$, EtOH, 100 °C

SCHEME 2

Although, the yield of the desired iso-HPMPA derivative **12** was lower compared to the procedure of Krečmerová et al.[10] (31% of diethyl ester of iso-HPMPA acetylated at C-2′ position), also their procedure suffers from

formation of a by-product (in this case a cyclic phosphonate) and acetylation of the reaction mixture was necessary to isolate the desired product. Reaction conditions of our procedure (alkylation/deprotection) were not further optimized.

Nucleophilic substitutions of bromine atom at the C-8 position of adenine with corresponding *O*- and *N*-nucleophiles were used for modifications of 9-{3-[(diisopropoxyphosphoryl)methoxy]-2-hydroxypropyl}adenine (**12**).

Bromination of the adenine derivative **12** with bromine in a mixture of dioxane and 10% aqueous solution of disodium hydrogen phosphate afforded 8-bromoadenine derivative **13** in 96% yield (Scheme 2). The attempt to brominate compound **11** under the same reaction conditions failed, probably because of the presence of the double bond at the aliphatic side chain and the reaction led to decomposition of the starting material.

Similarly as in the synthesis of 8,3′-*O*-anhydro HPMPA derivative **3** with annellated six-membered ring (Chart 1)[12b], 8-bromoadenine derivative **13** gave on treatment with 2 equivalents of sodium hydride in DMF 8,2′-*O*-anhydro derivative **14** with annellated five-membered cycle (Scheme 2). Free phosphonic acid **15**, the constitutional isomer of the compound **3**, was prepared in 26% yield by the standard procedure using TMSBr in acetonitrile followed by hydrolysis. The low yield of the compound **15** could be explained by susceptibility of the *O*-anhydro compounds to hydrolysis under acidic conditions[12b].

Treatment of the 8-bromoadenine derivative **13** with sodium acetate in acetic acid, followed by deprotection with TMSBr, gave 6-amino-7*H*-purine-8(9*H*)-one analogue **16** (Scheme 2). 8-(*N*-Methylamino)adenine derivative **17** and 8-(*N*,*N*-dimethylamino)adenine derivative **18** were obtained by reactions of the compound **13** with ethanolic solutions of either methylamine or dimethylamine, respectively, in an autoclave, followed by the deprotection of the phosphonate group.

In conclusion, synthesis of alkylating agent **9** was developed starting from allyl alcohol (**4**). Alkylation of adenine (**10**) with the tosylate **9**, followed by acid removal of the tetrahydro-2-pyranyl group, afforded 9-{3-[(diisopropoxyphosphoryl)methoxy]-2-hydroxypropyl}adenine (**12**) and the unsaturated derivative **11**. Bromination of the compound **12** gave 8-bromoadenine compound **13** quantitatively. Nucleophilic substitutions of the bromine atom of the compound **13**, followed by diester removal, afforded the free phoshonic acids **15–18**. Biological activities of the final compounds will be examined.

EXPERIMENTAL

Melting points were determined on a Büchi Melting Point B-545 apparatus and are uncorrected. NMR spectra (δ, ppm; *J*, Hz) were recorded on a Bruker Avance 500 (^{1}H at 500 MHz, ^{13}C at 125.8 MHz) in DMSO-d_6, or D_2O (with addition of NaOD to improve solubility). Chemical shifts were referenced to the solvent signal (DMSO-d_6 for ^{1}H NMR δ 2.5 ppm and for ^{13}C δ 39.7). Chemical shifts in D_2O were referenced to 1,4-dioxane for ^{1}H NMR δ 3.75 and for ^{13}C NMR δ 67.19. UV spectra were measured on DU Spectrophotometer with solutions in H_2O. Mass spectra were measured on a ZAB-EQ (VG Analytical) spectrometer using FAB (ionization by Xe, accelerating voltage 8 kV, glycerol matrix). The elemental analyses were obtained on a Perkin–Elmer CHN Analyzer 2400, Series II Sys (Perkin–Elmer). Column chromatography and thin-layer chromatography (TLC) were performed using Silica gel 60 (Fluka) and Silufol Silica gel 60 F254 foils (Merck), respectively. Solvents were evaporated at 2 kPa and bath temperature 30–60 °C. The compounds were dried at 2 kPa over P_2O_5.

Diisopropyl [(Prop-2-en-1-yloxy)methyl]phosphonate (**6**)[17]

Gaseous hydrogen chloride was introduced into the cooled (0 °C) mixture of allyl alcohol (**4**; 116.2 g, 2.0 mol) and formaldehyde (180.2 g, 6.0 mol) for 3 h. Then the upper organic layer was decanted off, dried ($CaCl_2$), and the drying agent was filtered off. Distillation of the residue afforded 3-(chloromethoxy)prop-1-ene[15] (**5**; 145.0 g, 68%) as a colorless oil, b.p. 108–110 °C.

A mixture of compound **5** (145.0 g, 1.4 mol) and triisopropyl phosphite (291.5 g, 1.4 mol) was refluxed at 120 °C for 7 h and distilled in vacuo affording compound **6** (155.9 g, 47%) as an oil, b.p. 90–92 °C at 198 kPa. MS (FAB), *m/z*: 237 $[M + H]^+$ (40), 153 $[M - 2\ (iPr) + H]^+$ (100).

Diisopropyl [(2-Hydroxy-3-tosyloxypropoxy)methyl]phosphonate (**8**)

A mixture of compound **6** (47.3 g, 0.2 mol), 50% aqueous ethanol (350 ml), $NaClO_3$ (59.6 g, 0.6 mol) and OsO_4 (0.20 g, 0.80 mmol) in CCl_4 (20 ml) was heated at 50 °C for 6 h and then stirred at room temperature overnight. Organic solvents were evaporated and the resulting solution was saturated with NaCl. The solution was extracted with diethyl ether (3 × 100 ml) and the organics were dried ($MgSO_4$). Filtration, evaporation of organics and distillation in vacuo gave compound **7** (19 g, 35%) as a colorless oil, b.p. 135–137 °C at 9.9 kPa. MS (FAB), *m/z*: 271 $[M + H]^+$ (100).

A solution of tosyl chloride (19 g, 0.10 mol) in pyridine (30 ml) was added dropwise with stirring to ice-cool solution of compound **7** (30 g, 0.11 mol) and DMAP (0.3 g) in pyridine (80 ml). The mixture was stirred at 0 °C for 3 h and set aside at room temperature overnight. Methanol (25 ml) was added and the mixture was stirred at room temperature for 1 h. The mixture was diluted with EtOAc (500 ml), washed with water (2 × 100 ml), and the organics were evaporated in vacuo. The residue was codistilled with toluene (3 × 50 ml) and dried over phosphorus pentoxide to give compound **8** (28 g, 60%) as a thick oil. ^{1}H NMR (DMSO-d_6): 1.19 d, 6 H, $J(CH_3,CH)$ = 6.1 (CH_3); 1.22 d, 6 H, $J(CH_3,CH)$ = 6.1 (CH_3); 2.42 s, 3 H (CH_3-arom); 3.40 dd, 1 H, J(3b,2) = 6.1, J(gem) = 10.0 (H-3b); 3.46 dd, 1 H, J(3a,2) = 5.1, J(gem) = 10.0 (H-3a); 3.69 d, 2 H, $J(P,CH)$ = 7.9 (PCH_2); 3.78 m, 1 H (H-2); 3.85 dd, 1 H, J(1b,2) = 6.5, J(gem) = 9.9 (H-1b); 3.98 dd, 1 H, J(1a,2) = 3.4, J(gem) = 9.9 (H-1a); 4.56 m, 2 H (POCH); 5.34 d, 1 H, J(OH,2) = 5.4 (OH); 7.48 d, 2 H, J = 8.3 (arom); 7.77 d, 2 H, J = 8.3

(arom). ^{13}C NMR (DMSO-d_6): 21.26 (CH_3-arom); 23.84 d, 2 C, J(P,C) = 4.9 (CH_3); 23.96 d, 2 C, J(P,C) = 3.9 (CH_3); 65.36 d, J(P,C) = 164.1 (PC); 67.01 (C-2); 70.29 d, 2 C, J(P,C) = 5.9 (POC); 72.05 (C-1); 73.19 d, J(P,C) = 10.7 (C-3); 127.79 (2 C, C-arom); 130.33 (2 C, C-arom), 132.43 (C-arom); 145.07 (C-arom).

Diisopropyl {[2-(Tetrahydro-2-pyranyl)oxy-3-tosyloxypropoxy]methyl}phosphonate (**9**)

A mixture of tosyl derivative **8** (28.0 g, 66.0 mmol), 3,4-dihydro-2*H*-pyran (12.3 g, 13.3 ml, 146.2 mmol) and HCl/DMF (4.5 mol/l, 2 ml) was stirred at room temperature for 4 h. The mixture was diluted with diethyl ether (150 ml) and washed (3 × 50 ml of water, 50 ml of 1 M $NaHCO_3$ aqueous solution, and 50 ml of water). The organic solvent was taken down in vacuo, the residue was codistilled with ethanol (3 × 50 ml) and dried over phosphorus pentoxide to give compound **9** (20.8 g, 62%) as a thick oil. ^{1}H NMR (DMSO-d_6): 1.19 d, 6 H, J(CH_3,CH) = 6.1 (CH_3); 1.22 d, 6 H, J(CH_3,CH) = 6.1 (CH_3); 1.53–1.89 m, 6 H (pyr); 2.41 s, 3 H (CH_3-arom); 3.53 dd, 1 H, J(3b,2) = 6.1, J(gem) = 10.0 (H-3b); 3.95 dd, 1 H, J(3a,2) = 5.1, J(gem) = 10.0 (H-3a); 3.61 m, 1 H (pyr); 3.69 d, 2 H, J(P,CH) = 7.9 (PCH_2); 3.74 m, 1 H (pyr); 4.35 dd, 1 H, J(1b,2) = 6.5, J(gem) = 9.9 (H-1b); 4.46 dd, 1 H, J(1a,2) = 3.4, J(gem) = 9.9 (H-1a); 4.68 m, 1 H (H-2); 4.60 m, 2 H (POCH); 4.76 m, 1 H (pyr); 7.46 d, 2 H, J = 8.3 (arom); 7.75 d, 2 H, J = 8.3 (arom).

Alkylation of Adenine with Tosylate **9**

A mixture of adenine (**10**; 5.0 g, 37.0 mmol), DBU (6.2 g, 6.1 ml, 40.7 mmol) and compound **9** (21 g, 41.3 mmol) in DMF (70 ml) was stirred at 110 °C for 22 h. The solvent was evaporated in vacuo, the residue codistilled with toluene (3 × 30 ml), and dissolved in chloroform (150 ml). The solution was washed with water (3 × 25 ml) and the organic solvent was taken down. The residue was heated in 1 M HCl (120 ml) at 60 °C for 2 h. The mixture was neutralized with 1 M NaOH and extracted with chloroform (3 × 50 ml). The extract was chromatographed on silica gel (10% MeOH in $CHCl_3$) and fractions were crystallized from EtOH to give compounds **11** (4.2 g, 31%) and **12** (1.6 g, 11%).

9-{3-[(Diisopropoxyphosphoryl)methoxy]prop-1-enyl}adenine (**11**). White crystals, m.p. 123–125 °C. MS (FAB), *m/z*: 370 $[M + H]^+$ (100). ^{1}H NMR (DMSO-d_6): 1.25 d, 6 H, J(CH_3,CH) = 6.1 (CH_3); 1.255 d, 6 H, J(CH_3,CH) = 6.1 (CH_3); 3.78 d, 2 H, J(P,CH) = 8.6 (PCH_2); 4.24 d, 2 H, J(3′,2′) = 6.4 (H-3′); 4.62 m, 2 H (POCH); 6.69 dt, 1 H, J(2′,3′) = 6.4, J(2′,1′) = 14.5 (H-2′); 7.31 d, 1 H, J(1′,2′) = 14.5 (H-1′); 7.38 brs, 2 H (NH_2), 8.19 and 8.48 s, 2 × 1 H (H-2 and H-8). ^{13}C NMR (DMSO-d_6): 23.88 d, 2 C, J(P,C) = 4.9 (CH_3); 23.99 d, 2 C, J(P,C) = 3.9 (CH_3); 46.50 (C-1′); 65.50 d, J(P,C) = 163.5 (PC); 67.68 (C-2′); 70.42 d, 2 C, J(P,C) = 6.4 (POC); 74.85 d, J(P,C) = 10.3 (C-3′); 118.78 (C-5); 141.74 (C-8); 149.86 (C-4); 152.46 (C-2); 156.11 (C-6). For $C_{15}H_{26}N_5O_5P$ (387.4) calculated: 46.51% C, 6.76% H, 18.08% N, 8.00% P; found: 46.42% C, 6.78% H, 17.82% N, 8.10% P.

(R,S)-9-{3-[(Diisopropoxyphosphoryl)methoxy]-2-hydroxypropyl}adenine (**12**). White crystals, m.p. 125–127 °C. MS (FAB), *m/z*: 388 $[M + H]^+$ (100). ^{1}H NMR (DMSO-d_6): 1.24 d, 6 H, J(CH_3,CH) = 6.2 (CH_3); 1.25 d, 6 H, J(CH_3,CH) = 6.2 (CH_3); 3.49 dd, 1 H, J(3′b,2′) = 5.2, J(gem) = 12.4 (H-3′b); 3.52 dd, 1 H, J(3′a,2′) = 5.0, J(gem) = 12.4 (H-3′a); 3.80 d, 2 H, J(P,CH) = 7.8 (PCH_2); 4.01 m, 1 H (H-2′); 4.04 dd, 1 H, J(1′b,2′) = 8.1, J(gem) = 12.4 (H-1′b); 4.26 dd, 1 H, J(1′a,2′) = 2.2, J(gem) = 12.4 (H-1′a); 4.61 m, 2 H (POCH); 5.34 d, 1 H, J(OH,2′) = 5.1 (OH); 7.21 brs, 2 H (NH_2); 8.05 and 8.13 s, 2 × 1 H (H-2 and H-8). ^{13}C NMR

(DMSO-d_6): 23.88 d, 2 C, J(P,C) = 4.9 (CH_3); 23.99 d, 2 C, J(P,C) = 3.9 (CH_3); 46.50 (C-1′); 65.50 d, J(P,C) = 163.5 (PC); 67.68 (C-2′); 70.42 d, 2 C, J(P,C) = 6.4 (POC); 74.85 d, J(P,C) = 10.3 (C-3′); 118.78 (C-5); 141.74 (C-8); 149.86 (C-4); 152.46 (C-2); 156.11 (C-6). For $C_{15}H_{26}N_5O_5P$ (387.4) calculated: 46.51% C, 6.76% H, 18.08% N, 8.00% P; found: 46.42% C, 6.78% H, 17.82% N, 8.10% P.

(*R,S*)-8-Bromo-9-{3-[(diisopropoxyphosphoryl)methoxy]-2-hydroxypropyl}adenine (**13**)

Bromine (0.10 ml, 0.31 g, 3.9 mmol) was added to a suspension of compound **12** (0.80 g, 2.1 mmol) in a mixture of dioxane (40 ml) and 10% aqueous solution of sodium hydrogenphosphate (25 ml) and the mixture was stirred at room temperature for 15 h. A concentrated solution of sodium hydrogensulfite was added until the mixture became colorless and the product was taken up in chloroform (5 × 30 ml). The chloroform extract was dried over anhydrous magnesium sulfate, filtered, and the solvent was evaporated. Purification on a silica gel plate (15% MeOH in $CHCl_3$) and crystallization (EtOAc) afforded compound **13** (0.92 g, 96%) as yellowish crystals, m.p. 138–140 °C. MS (FAB), *m/z*: 466/468 $[M + H]^+$ (100). ^{1}H NMR (DMSO-d_6): 1.25 d, 12 H, J(CH_3,CH) = 6.1 (CH_3); 3.55 dd, 1 H, J(3′b,2′) = 5.1, J(gem) = 10.1 (H-3′b); 3.59 dd, 1 H, J(3′a,2′) = 5.1, J(gem) = 10.1 (H-3′a); 3.78 dd, 1 H, J(P,CH) = 7.7, J(gem) = 13.9 (PCH_2); 3.82 dd, 1 H, J(P,CH) = 8.8, J(gem) = 13.9 (PCH_2); 4.07 dd, 1 H, J(1′b,2′) = 8.1, J(gem) = 12.8 (H-1′b); 4.13 m, 1 H (H-2′); 4.17 dd, 1 H, J(1′a,2′) = 3.4, J(gem) = 12.8 (H-1′a); 4.61 m, 2 H (POCH); 4.96 d, 1 H, J(OH,2′) = 5.2 (OH); 7.36 brs, 2 H (NH_2); 8.12 s, 1 H (H-2). ^{13}C NMR (DMSO-d_6): 23.96 d, 2 C, J(P,C) = 4.5 (CH_3); 24.05 d, 2 C, J(P,C) = 3.7 (CH_3); 47.68 (C-1′); 65.50 d, J(P,C) = 163.6 (PC); 67.30 (C-2′); 70.38 d, J(P,C) = 6.3 (POC); 70.40 d, J(P,C) = 6.3 (POC); 75.00 d, J(P,C) = 10.2 (C-3′); 119.20 (C-5); 127.39 (C-8); 151.21 (C-4); 152.79 (C-2); 154.91 (C-6).

Diisopropyl (*R,S*)-{[(4-Amino-7,8-dihydro-[1,3]oxazolo[3,2-*e*]purin-7-yl)methoxy]methyl}-phosphonate (**14**)

A mixture of compound **13** (0.6 g, 1.3 mmol) and 60% sodium hydride suspension (100 mg, 2.6 mmol) in DMF (20 ml) was stirred at room temperature for 1 h. The mixture was neutralized (AcOH) and taken down in vacuo. The residue was codistilled with toluene (2 × 10 ml) and purified on a silica gel plate (15% MeOH in $CHCl_3$). Crystallization (EtOAc) afforded compound **14** (0.36 g, 72%) as white crystals, m.p. 150–152 °C. MS (FAB), *m/z*: 386 $[M + H]^+$ (30), 302 $[M - 2\ (iPr) + H]^+$ (100). ^{1}H NMR (DMSO-d_6): 1.14 d, 6 H, J(CH_3,CH) = 6.1 (CH_3); 1.17 d, 3 H, J(CH_3,CH) = 6.1 (CH_3); 1.18 d, 3 H, J(CH_3,CH) = 6.1 (CH_3); 3.84 dd, 1 H, J(P,CH) = 9.2, J(gem) = 13.9 (PCH_2); 3.87 dd, 1 H, J(P,CH) = 9.2, J(gem) = 13.9 (PCH_2); 3.88 dd, 1 H, J(3′b,2′) = 4.4, J(gem) = 11.6 (H-3′b); 3.98 dd, 1 H, J(3′a,2′) = 2.6, J(gem) = 11.6 (H-3′a); 4.07 dd, 1 H, J(1′b,2′) = 6.0, J(gem) = 9.4 (H-1′b); 4.37 dd, 1 H, J(1′a,2′) = 8.8, J(gem) = 9.4 (H-1′a); 4.50 m, 2 H (POCH); 5.63 m, 1 H (H-2′); 6.74 brs, 2 H (NH_2), 7.98 s, 1 H (H-2). ^{13}C NMR (DMSO-d_6): 23.76 d, 2 C, J(P,C) = 3.9 (CH_3); 23.91 d, 2 C, J(P,C) = 3.9 (CH_3); 42.13 (C-1′); 65.39 d, J(P,C) = 164.1 (PC); 70.44 d, J(P,C) = 6.8 (POC); 70.47 d, J(P,C) = 6.8 (POC); 72.60 d, J(P,C) = 10.7 (C-3′); 86.70 (C-2′); 120.34 (C-5); 147.10 (C-4); 150.54 (C-2); 154.00 (C-6); 160.25 (C-8). For $C_{15}H_{24}N_5O_5P$ (385.4) calculated: 46.75% C, 6.28% H, 18.17% N, 8.04% P; found: 46.46% C, 6.28% H, 17.82% N, 7.97% P.

(*R*,*S*)-{[(4-Amino-7,8-dihydro[1,3]oxazolo[3,2-*e*]purin-7-yl)methoxy]methyl}-phosphonic Acid (**15**)

Method A [19]: A mixture of compound **14** (386 mg, 1.0 mmol) and TMSBr (1.0 ml) in acetonitrile (5 ml) was stirred overnight at ambient temperature, then evaporated and codistilled with acetonitrile (10 ml). The residue was dissolved in water and alkalized with aqueous ammonia. The mixture was evaporated in vacuo and the residue was deionized on a Dowex 50X8 (H^+) column (50 ml) under standard conditions (washing with water and elution with 2.5% aqueous ammonia). The product was purified on a Dowex 1X2 (acetate) column by elution with linear gradient of acetic acid (0–0.5 mol/l, 1 l each). The UV-absorbing fractions were evaporated in vacuo and the residue was crystallized to afford compound **15** (79 mg, 26%) as white crystals (H_2O), m.p. > 270 °C (dec.), E_{Up} = 0.80. MS (FAB), *m/z*: 302 $[M + H]^+$ (100). ^{1}H NMR (D_2O): 3.61 d, 2 H, *J*(P,CH) = 8.3 (PCH_2); 4.01 dd, 1 H, *J*(3′b,2′) = 6.0, *J*(gem) = 12.0 (H-3′b); 4.06 dd, 1 H, *J*(3′a,2′) = 3.3, *J*(gem) = 12.0 (H-3′a); 4.24 dd, 1 H, *J*(1′b,2′) = 6.9, *J*(gem) = 9.5 (H-1′b); 4.47 dd, 1 H, *J*(1′a,2′) = 8.9, *J*(gem) = 9.5 (H-1′a); 5.81 m, 1 H (H-2′); 8.09 s, 1 H (H-2). ^{13}C NMR (D_2O): 45.43 (C-1′); 72.53 d, *J*(P,C) = 149.4 (PC); 74.86 d, *J*(P,C) = 8.8 (C-3′); 91.40 (C-2′); 122.51 (C-5); 149.01 (C-4); 153.13 (C-2); 155.82 (C-6); 163.91 (C-8). For $C_9H_{12}N_5O_5P$ (301.2) calculated: 35.89% C, 4.02% H, 23.25% N, 10.28% P; found: 35.76% C, 4.15% H, 22.99% N, 10.17% P. UV, λ_{max} (ε_{max}): (pH 2) 262 (12 100); (pH 12) 262 (12 400).

(*R*,*S*)-6-Amino-9-[2-hydroxy-3-(phosphonomethoxy)propyl]-7H-purin-8(9*H*)-one (**16**)

A mixture of compound **13** (933 mg, 2 mmol), acetic acid (15 ml) and sodium acetate (1.31 g, 16 mmol) was refluxed for 8 h. Volatiles were evaporated in vacuo and the residue was codistilled with water (3 × 10 ml). The residue was stirred with sodium methoxide (10 ml, 125 mmol) at 50 °C for 2 h, neutralized with HCl, and volatiles were evaporated in vacuo. The residue was deionized on a Dowex 50X8 (H^+) column (50 ml, elution with water followed by 2.5% aqueous ammonia). The UV-absorbing ammonia eluate was collected and evaporated in vacuo. The crude intermediate was dried over phosphorus pentoxide overnight and then treated with TMSBr under standard conditions (method *A*) to give compound **16** (237 mg, 37%) as yellowish crystals (H_2O), m.p. 275 °C, E_{Up} = 0.81. MS (FAB), *m/z*: 320 $[M + H]^+$ (100). ^{1}H NMR (D_2O): 3.47 dd, 1 H, *J*(P,CHb) = 8.9, *J*(gem) = 12.6 (PCHb); 3.52 dd, 1 H, *J*(P,CHa) = 8.7, *J*(gem) = 12.6 (PCHa); 3.54 dd, 1 H, *J*(3′b,2′) = 7.2, *J*(gem) = 10.8 (H-3′b); 3.60 dd, 1 H, *J*(3′a,2′) = 3.1, *J*(gem) = 10.8 (H-3′a); 3.89 dd, 1 H, *J*(1′b,2) = 5.9, *J*(gem) = 14.4 (H-1′b); 3.95 dd, 1 H, *J*(1′a,2′) = 7.3, *J*(gem) = 14.4 (H-1′a); 4.24 m, 1 H (H-2′); 7.96 s, 1 H (H-2). ^{13}C NMR (D_2O): 46.57 (C-1′); 70.90 (C-2′); 72.32 d, *J*(P,C) = 150.3 (PC); 76.48 d, *J*(P,C) = 11.3 (C-3′); 103.35 (C-5); 146.59 and 146.60 (C-4 and C-6); 150.47 (C-2); 151.52 (C-8). HR MS (FAB) for $C_9H_{15}N_5O_6P$ $[M + H]$ calculated 320.0760, found 320.0761. UV, λ_{max} (ε_{max}): (pH 2) 281 (10 500); (pH 7) 272 (12 300); (pH 12) 281 (13 900).

(*R*,*S*)-9-[2-Hydroxy-3-(phosphonomethoxy)propyl]-8-(*N*-methylamino)adenine (**17**)

Method B [12d]: A mixture of compound **13** (933 mg, 2 mmol) in ethanolic methylamine solution (33%, 30 ml) was heated in a steel autoclave at 100 °C for 15 h. Then the solvents were evaporated, the residue was codistilled with ethanol (2 × 15 ml) and dried over phosphorus pentoxide overnight. The crude intermediate was treated with TMSBr under standard conditions (method *A*) to give compound **17** (299 mg, 45%) as white crystals (H_2O), m.p.

217–219 °C, E_{Up} = 0.75. MS (FAB), *m/z*: 333 $[M + H]^+$ (100). ^{1}H NMR (D_2O + NaOD): 2.98 s, 3 H (NCH_3); 3.49 dd, 1 H, *J*(3′b,2′) = 5.7, *J*(gem) = 10.5 (H-3′b); 3.50 dd, 1 H, *J*(P,CHb) = 8.8, *J*(gem) = 12.4 (PCHb); 3.53 dd, 1 H, *J*(P,CHa) = 8.9, *J*(gem) = 12.4 (PCHa); 3.59 dd, 1 H, *J*(3′a,2′) = 3.8, *J*(gem) = 10.5 (H-3′a); 3.92 dd, 1 H, *J*(1′b,2′) = 7.1, *J*(gem) = 15.0 (H-1′b); 3.96 dd, 1 H, *J*(1′a,2′) = 5.1, *J*(gem) = 15.0 (H-1′a); 4.13 m, 1 H (H-2′); 7.92 s, 1 H (H-2). ^{13}C NMR (D_2O + NaOD): 31.43 (NCH_3), 46.66 (C-1′); 70.85 (C-2′); 72.27 d, *J*(P,C) = 150.5 (PC); 76.43 d, *J*(P,C) = 11.3 (C-3′); 118.90 (C-5); 151.53 (C-2); 152.59 (C-4); 153.55 (C-6); 157.07 (C-8). For $C_{10}H_{17}N_6O_5P$ (332.3) calculated: 36.15% C, 5.16% H, 25.29% N, 9.32% P; found: 35.94% C, 5.24% H, 24.96% N, 9.05% P. UV, λ_{max} (ε_{max}): (pH 2) 275 (11 400); (pH 12) 279 (13 900).

(*R*,*S*)-8-(*N*,*N*-Dimethylamino)-9-[2-hydroxy-3-(phosphonomethoxy)propyl]adenine (**18**)

Treatment of compound **13** (933 mg, 2 mmol) with ethanolic dimethylamine solution (33%, 30 ml) by method *B*, followed by deprotection (method *A*) gave compound **18** (270 mg, 39%) as white crystals (H_2O), m.p. 250 °C, E_{Up} = 0.77. MS (FAB), *m/z*: 347 $[M + H]^+$ (100). ^{1}H NMR (D_2O + NaOD): 3.04 s, 6 H (NCH_3); 3.51 dd, 1 H, *J*(P,CHb) = 8.9, *J*(gem) = 12.4 (PCHb); 3.54 dd, 1 H, *J*(P,CHa) = 8.9, *J*(gem) = 12.4 (PCHa); 3.56 dd, 1 H, *J*(3′b,2′) = 6.5, *J*(gem) = 10.6 (H-3′b); 3.64 dd, 1 H, *J*(3′a,2′) = 3.3, *J*(gem) = 10.6 (H-3′a); 4.19 dd, 1 H, *J*(1′b,2′) = 4.8, *J*(gem) = 15.0 (H-1′b); 4.24 dd, 1 H, *J*(1′a,2′) = 8.4, *J*(gem) = 15.0 (H-1′a); 4.36 m, 1 H (H-2′); 8.04 s, 1 H (H-2). ^{13}C NMR (D_2O + NaOD): 43.82 (2 C, NCH_3), 48.90 (C-1′); 70.00 (C-2′); 72.21 d, *J*(P,C) = 150.4 (PC); 76.80 d, *J*(P,C) = 11.3 (C-3′); 118.75 (C-5); 152.80 (C-2); 153.38 (C-4); 155.04 (C-6); 160.01 (C-8). For $C_{11}H_{19}N_6O_5P$ (346.3) calculated: 38.15% C, 5.53% H, 24.27% N, 8.94% P; found: 37.96% C, 5.67% H, 24.01% N, 8.92% P. UV, λ_{max} (ε_{max}): (pH 2) 282 (15 800); (pH 12) 278 (16 200).

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