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Synthesis of branched 9-[2-(2-phosphonoethoxy)ethyl]purines as a new class of acyclic nucleoside phosphonates which inhibit *Plasmodium falciparum* hypoxanthine–guanine–xanthine phosphoribosyltransferase

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ABSTRACT

The malarial parasite *Plasmodium falciparum* (*Pf*) lacks the de novo pathway and relies on the salvage enzyme, hypoxanthine–guanine–xanthine phosphoribosyltransferase (HGXPRT), for the synthesis of the 6-oxopurine nucleoside monophosphates. Specific acyclic nucleoside phosphonates (ANPs) inhibit *Pf*HGXPRT and possess anti-plasmodial activity. Two series of novel branched ANPs derived from 9-[2-(2-phosphonoethoxy)ethyl]purines were synthesized to investigate their inhibition of *Pf*HGXPRT and human HGPRT. The best inhibitor of *Pf*HGXPRT has a K_i of 1 μM. The data showed that both the position and nature of the hydrophobic substituent change the potency and selectivity of the ANPs.

1. Introduction

Malaria is one of the most widespread infectious diseases in the world. It is caused by protozoan parasites of the genus *Plasmodium*. The most lethal of the strains that infect humans is *Plasmodium falciparum* (*Pf*).[1] Widespread resistance to most of the commonly used anti-malarial drugs is emerging. Therefore, replacement chemotherapies are urgently needed.[2] Since many of the metabolic pathways present in the parasite are identical to those of its host cells, the identification of new targets and the design of specific inhibitors are challenging.

One pathway where humans and protozoan parasites differ significantly is purine metabolism.[3,4] Mammals are able to synthesize 6-oxopurine nucleoside monophosphates by the de novo pathway as well as by salvage of preformed purine bases. In contrast, parasites can only produce their 6-oxopurine nucleoside monophosphates (necessary for many cellular processes) from purine bases transported from their host cell.[5] Hypoxanthine–guanine–xanthine phosphoribosyltransferase (HGXPRT) is a key enzyme of purine salvage pathway and its activity is essential for the replication and survival of the parasite[6] while human cells are not as dependent on the activity of the corresponding human enzyme—hypoxanthine–guanine phosphoribosyltransferase (HGPRT). *Pf*HGXPRT is, therefore, recognized as a target for the design of anti-malarial drugs.[7,8] The reaction catalysed by these 6-oxopurine phosphoribosyltransferases is shown in Figure 1.

Acyclic nucleoside phosphonates (ANPs) where the purine base is attached to a phosphonate moiety via a linker are structural analogs of the nucleoside monophosphate product of the reaction.[9] The ANPs are the first inhibitors that have been demonstrated to selectively discriminate between human HGPRT and *Pf*HGXPRT. Compounds with the phosphonoethoxyethyl (PEE) moiety attached to the purine base inhibit *Pf*HGXPRT with K_i values of 0.1 (PEEG) and 0.3 (PEEHx) μM, respectively.[10] However, they are weaker inhibitors of the human enzyme and the ratio in favour of *Pf*HGXPRT ($K_{i\ (human)}/K_{i\ (Pf)}$) is of the order of a factor of 10. These two compounds (Fig. 2) have been crystallized in complex with human HGPRT showing the specific interactions between the ANPs and the amino acid residues located at the active site.[10]

Acyclic nucleoside phosphonates (ANPs) are currently in use as anti-viral agents.[11,12] Such nucleotide analogues are excellent

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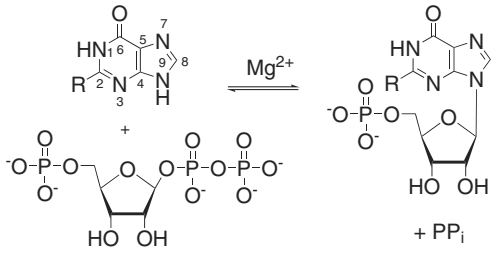


Figure 1. The reaction catalysed by the 6-oxopurine phosphoribosyltransferases. R = NH_2 (guanine), R = H (hypoxanthine), R = OH (xanthine).

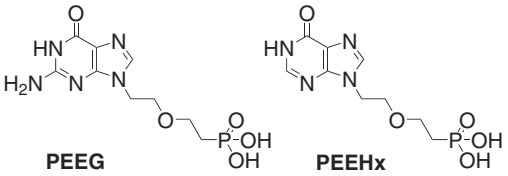


Figure 2. Inhibitors of HG(X)PRT. The structure of two ANPs with the PEE moiety attached to the purine base.

templates for drug design because of the absence of a labile glycosidic bond and stability of phosphonate moiety compared with the phosphate ester bond that can be easily hydrolyzed. The presence of a phosphonate group in ANPs is responsible for their highly polar character and deprotonation at physiological pH. ANPs can be transformed to pro-drugs to improve their pharmacological properties and to increase their ability to cross cell membranes.[13,14] Specific ANPs which are good inhibitors of *Pf*HGXPRT possess anti-plasmodial activity with IC_{50} values of 1 μM.[9,10] Their pharmacokinetic properties and their low cytotoxicity towards mammalian cells make them prime candidates for the development of chemotherapeutics that exert even higher toxicity towards *P. falciparum*.

Herein, we report the synthesis of a new series of ANP derivatives based on the phosphonoethoxyethyl (PEE) ANPs. These compounds were designed to investigate the effect of branching of phosphonoethoxyethyl chain on their ability to inhibit *Pf*HGXPRT or human HGPRT. Substituents were added either to the carbon atom

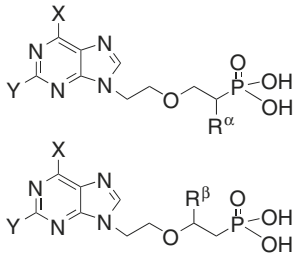


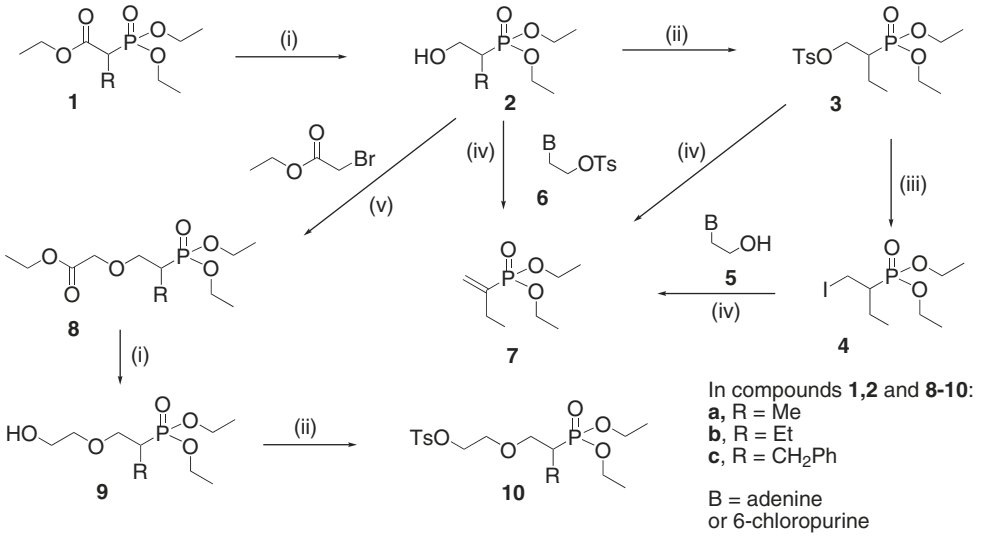
Figure 3. Structures of the target ANPs with α- and β-branched chain.

tached directly to phosphorus atom (α-branched derivatives) or to the adjacent carbon (β-branched derivatives) (Fig. 3). Methyl, ethyl and benzyl were selected as model substituents of different size. These compounds were tested for their cytotoxicity in mammalian cells and the influence of the addition of the different hydrophobic groups to the PEE moiety on the inhibition of human HGPRT or *Pf*HGXPRT.

2. Chemistry

For the efficient synthesis of the branched-PEE acyclic nucleoside phosphonates, the retrosynthetic disconnection of the ether bond was an obvious choice. All compounds were planned to be prepared in racemic form for preliminary screening. Although the methodology for ANPs synthesis is well established, the preparation of compounds containing branched linkers between the heterocyclic base and the phosphonate moiety is often complicated by elimination reactions. Thus, development of synthetic procedures for this new class of ANPs was necessary.

Commercially available triethyl phosphonoacetate can be used as a starting material for the phosphonate-containing part of the α-branched PEE chain. This compound can be easily alkylated to introduce any desired substituent into chain. This known reaction[15] was applied to prepare benzyl derivative **1c**. Phosphonates **1a** and **1b** with methyl and ethyl substituent are commercially available, but can be also synthesized in the same way. Very clean and easy to perform reduction[16] of carboxylic acid ester group of phosphonates **1** by solution of borane in THF (Scheme 1) afforded alcohols **2a–c** in good yields. The terminal hydroxyl group of ethyl



Scheme 1. Reagents: (i) BH_3·THF; (ii) TsCl, CH_2Cl_2, DMAP, Et_3N; (iii) NaI, acteone; (iv) base (NaH, Et_3N, BuLi, DBU or *t*BUOK), THF or DMF; (v) NaH, THF, Et_3N.

derivative **2b** (chosen as model representative) was tosylated (compound **3**) and transformed to the iodo derivative **4** (Scheme 1, Supplementary data).

9-Hydroxyethyladenine or 9-hydroxyethylchloropurine (**5**), were prepared according to known procedure[17] using the reaction of the purine base with ethylene carbonate in the presence of catalytic amount of NaOH. The hydroxyl group was tosylated to form the derivatives **6** (Supplementary data).

Unfortunately, all our attempts to form the ether bond by the alkylation of hydroxyethylpurines **5** with tosyl derivative **3** or iodo derivative **4** failed. Also unsuccessful was the reversed alkylation of alcohols **2** with tosylates **6**. Under all conditions (DMF, THF as solvent; NaH, Et_3N, BuLi, DBU, *t*BuOK as base) branched phosphonates **2**–**4** strongly preferred elimination and so as a consequence only undesired known[21] substituted vinyl phosphonate **7** was isolated.

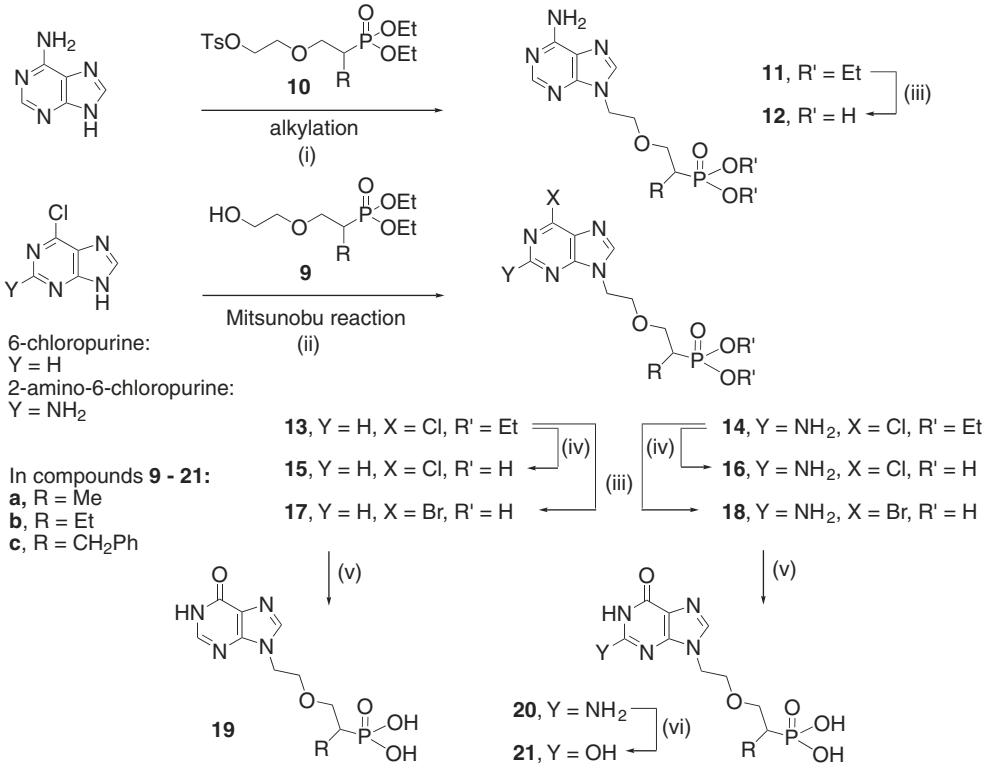
The strategy for ether bond formation was thus changed. It was clear, that only a strong alkylating agent and mild reaction conditions could be used. To meet these conditions, mixture of NaH, Et_3N and dry THF was cooled to −40 °C under argon atmosphere. Alcohol **2** was added followed, after 30 min, by ethyl bromoacetate. Ethyl (alkoxy)acetate moiety in the resulting key intermediates **8a**–**c** can serve as latent hydroxyethyl group. These esters **8** were reduced by borane in THF to obtain desired hydroxy derivatives **9** with the already built-in ether bond giving 58–77% yield in the two steps. Their tosylation under standard conditions afforded tosylates **10** in moderate yields 50–60%.

Two methods were used to introduce the α-branched phosphonate chain into the 9-position of purine base (Scheme 2). Standard alkylation of adenine by tosylderivatives **10** using Cs_2CO_3 as a base afforded ANP diethyl esters **11a**–**c** in 63–70% yield. After subsequent treatment with bromotrimethylsilane followed by hydrolysis the free phosphonic acids **12a**–**c** were isolated. In contrast, 6-chloropurine and 2-amino-6-chloropurine were alkylated by alcohols **9** under Mitsunobu conditions to form esters **13a**–**c** and **14a**–**c**, respectively. As expected, in this case yields were lower (34–68%), but the inefficient alcohol-tosylation step was skipped.

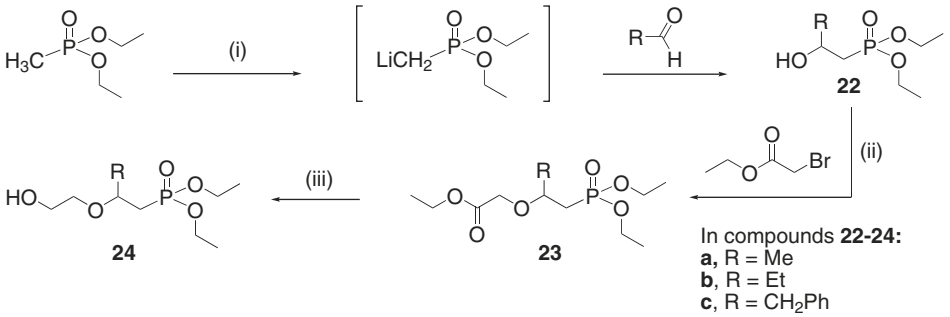
As suitable starting material for the synthesis of β-branched series, diethyl 2-hydroxyalkylphosphonates **22** were prepared according to an established protocol.[18] The same substituents (methyl, ethyl and benzyl) as in the case of α-branched ANPs were introduced to the β-position. Starting from commercially available diethyl methylphosphonate (Scheme 3), corresponding aldehydes were reacted with in situ preformed lithiomethylphosphonate intermediate. For the elongation of these β-branched alcohols **22**, the same sequence of alkylation and reduction step as for α-branched derivatives was applied. The resulting alcohols **24** were introduced under Mitsunobu conditions into 9-position of purine bases (Scheme 4) to form ANP diethyl esters **25a**–**c** and **26a**–**c**.

Further procedures were analogous for both series of substituted PEE compounds (Schemes 2 and 4). Cleavage of the esters **13**, **14**, **25** and **26** under standard conditions (Me_3SiBr/acetonitrile) led to the simultaneous exchange of chlorine in the 6-position on purine to bromine (derivatives **17**, **18**, **29** and **30**) as a result of contamination of bromotrimethylsilane by HBr. When modified reaction conditions in the presence of 2,6-lutidine were used, the expected chloro derivatives **15a**,**c**, **16a**,**c**, **27a**–**c** and **28a**–**c** were obtained. For further modification of the purine bases, routine procedures were applied (Schemes 2 and 4). Hydrolysis of halogen in 6-position of purine derivatives gave finally target hypoxanthine and guanine ANPs (**19a**–**c**, **20a**–**c**, **31a**–**c** and **32a**–**c**). Diazotation of the guanine compounds **20** and **32** led to xanthine phosphonic acids **21a**–**c** and **33a**–**c**.

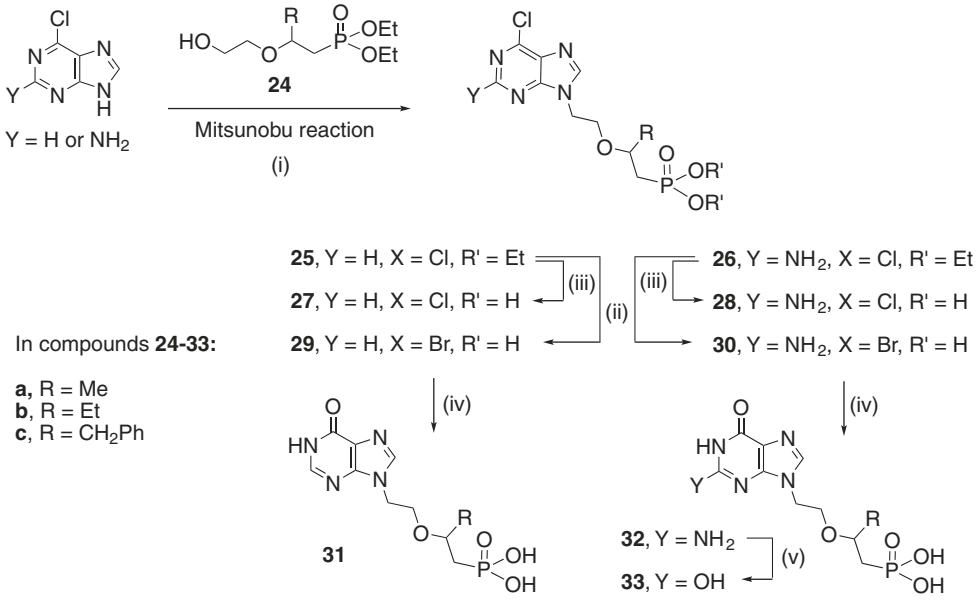
The title branched 9-(phosphonoethoxyethyl)purines **12a**–**c**, **15a**,**c**, **16a**,**c**, **17a**–**c**–**21a**–**c** and **27a**–**c**–**33a**–**c** (43 derivatives) were tested on their in vitro inhibition of the cell growth in mouse leukemia L1210 cells, humanT-lymphoblastoid CCRF-CEM cell line,



Scheme 2. Reagents: (i) Cs_2CO_3, DMF; (ii) PPh_3, DIAD, THF; (iii) CH_3CN, Me_3SiBr; (iv) CH_3CN, 2,6-lutidine, Me_3SiBr; (v) TFA/H_2O; (vi) $NaNO_2$, HCL/H_2O.



Scheme 3. Reagents: (i) *n*BuLi, THF; (ii) NaH, Et_3N, THF; (iii) BH_3·THF.



Scheme 4. Reagents: (i) PPh_3, DIAD, THF; (ii) CH_3CN, Me_3SiBr; (iii) CH_3CN, 2,6-lutidine, Me_3SiBr; (iv) TFA/H_2O; (v) $NaNO_2$, HCL/H_2O.

human promyelocytic leukemia HL-60 cells and human cervix carcinoma HeLa S3 cells (Dr. I. Votruba, IOCB). None of the compounds exhibited considerable cytostatic activity or cytotoxicity.

3. Inhibition of *Pf*HGXPRT and human HGPRT by the α- and β-branched ANPs

3.1. α-Branched chain ANPs

The α-branched derivatives either do not inhibit *Pf*HGXPRT or are very weak inhibitors at the concentration range used in the assay (60–100 μM). Similarly, they are also weak inhibitors of human HGPRT. The crystal structure[10] of human HGPRT in complex with PEEG and PEEHx, where a proton replaces the hydrophobic groups, suggests that steric hindrance is the primary reason for the increased K_i values (Fig. 4).

In the human structure, the carbon atom attached to phosphonate group of PEEG is located only 3.7 Å from the OG1 atom of T141 and 3.5 Å from the carbonyl oxygen of I135. Thus, there would be insufficient space to accommodate the methyl or ethyl groups and especially the bulky benzyl group. For such compounds to bind, the loop which encircles the 5′-phosphate binding pocket (encompassing residues 133–141) would have to change conformation. Thus, if the compound bound at all, fewer interactions between the phosphonate oxygens and the 5′-phosphate binding

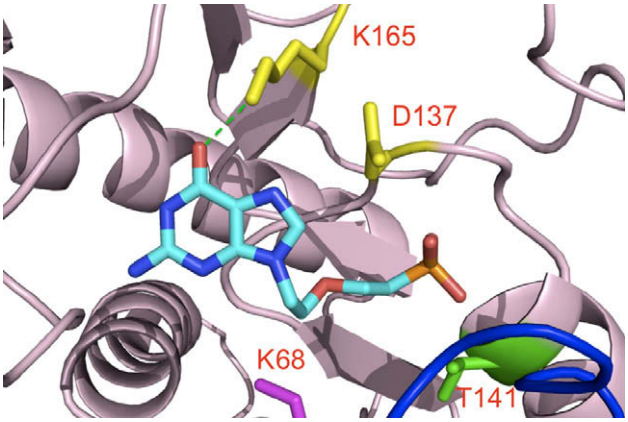


Figure 4. PEEG bound in the active site of human HGPRT.[10] The dark blue coil shows the location of the large mobile loop (residues 100–117) that partially covers the active site when this ANP binds.

pocket residues would occur. The amino acid sequence identity between human HGPRT and *Pf*HGXPRT is completely conserved in this region suggesting that the bonds between the phosphonate oxygens and the polypeptide should be identical in both enzymes. The inability of the phosphonate group in α-branched ANPs to bind

Table 1
Comparison of the K_i values for the β-branched chain ANPs with human HGPRT and *Pf* HGXPRT

Purine base	Substituent -R	Compound	K_i (μM) human HGPRT	K_i (μM) *Pf*HGXPRT	Ratio K_i (hu)/K_i (*Pf*)
Guanine	–H[a]	PEEG	1	0.1	10
	$-CH_3$	**32a**	0.1	1	0.1
	$-CH_2CH_3$	**32b**	0.3	5	0.06
Hypoxanthine	–H[a]	PEEHx	3.6	0.3	12
	$-CH_3$	**31a**	2	10	0.2
	$-CH_2CH_3$	**31b**	10	69	0.15

[a] Ref. 10.

makes these compounds extremely weak inhibitors of human HGPRT and *Pf*HGXPRT.

3.2. β-branched chain ANPs

When the base is either guanine or hypoxanthine, the β-branched derivatives inhibit both enzymes (Table 1). Changing the base to xanthine, adenine, 6-bromopurine, 2-amino-6-bromopurine, 6-chloropurine or 2-amino-6-chloropurine weakens the inhibition (K_i ranges between 50 and 200 μM). The inhibition is so weak that it is relatively independent of whether the attachment is a proton, methyl or ethyl group. For the human enzyme, with guanine as the base, the addition of the methyl or ethyl group allows these compounds to bind more tightly than when only a proton is attached. When hypoxanthine is the base, however, there is little difference in the calculated K_i value. The much lower value for the inhibition with guanine as the base may be a reflection of the fact that human HGPRT prefers guanine in terms of K_m (*cf.* 1.9 with 3.4 μM). Both enzymes prefer a methyl compared to an ethyl group because there is less space in the active site to accommodate the larger attachment. This argument is reinforced by the fact that when the larger benzyl group is attached to the linker, the resulting ANPs, effectively show no binding.

The β-branched ANP derivatives bind more weakly to *Pf*HGXPRT compared to human HGPRT. This may be because the large mobile loop is more firmly closed when PEEG and PEEHx bind to *Pf*HGXPRT compared with the branched chain ANPs. Thus, for *Pf*HGXPRT these hydrophobic additions increase the K_i value because the extra space they occupy does not allow the mobile loop to close as firmly.

4. Conclusions and perspectives

An efficient methodology for the synthesis of a novel class of α- and β-branched acyclic nucleoside phosphonates has been developed and a series of 43 derivatives synthesized. Attachment of a methyl or ethyl group to the phosphonate carbon changes the selectivity ratio ($K_{i\ (hu)}/K_{i\ (Pf)}$) from 10 (when the PEE moiety alone links guanine to the phosphonate group) to 0.1. Thus, the influence of these ANPs is not only in affinity but also in selectivity. These new results contribute to extensive SAR project that provide a focus for the development of new anti-malarials based on the ANP scaffold.

5. Experimental

Unless otherwise stated, solvents were evaporated at 40 °C/2 kPa, and the compounds were dried over P_2O_5 at 2 kPa. Preparative TLC was carried out on 40 × 17 × 0.4 cm loose layers of silica gel containing a UV indicator. NMR spectra were recorded on Bruker Avance 500 (^{1}H at 500 MHz, ^{13}C at 125.8 MHz) and Bruker Avance 400 (^{1}H at 400 MHz, ^{13}C at 100.6 MHz) spectrometers with TMS as internal standard or referenced to the residual solvent signal. Mass spectra were measured on a ZAB-EQ (VG Analytical) spectrometer. The chemicals were obtained from commercial sources (Sigma–Aldrich) or prepared according to the published procedures. Dimethylformamide and acetonitrile were distilled from P_2O_5 and stored over molecular sieves (4 Å). THF was distilled from sodium/benzophenone under argon. Preparative HPLC purifications were performed on columns packed with 7 μm C18 reversed phase resin (Waters Delta 600 chromatograph column), 17 × 250 mm; in ca. 200 mg batches of mixtures using gradient MeOH/H_2O as eluent. Deionisation was performed on Dowex 50 × 8 (H^+-form) columns by the following procedure: after application of crude product the column was washed with water until the UV absorption dropped. Thereafter, the column was eluted with 2.5% aqueous NH_3. Chromatography on Dowex 1 × 2 (acetate form) was as follows: after application of the aqueous solution of the crude product onto the column, it was washed with water until the UV absorption dropped. The column was then eluted with a gradient of dilute acetic or formic acid (0–1 M). All tested ANPs were characterized by ^{1}H NMR, ^{13}C NMR and mass spectrometry. The purity of the compounds was determined by combustion elemental analysis (C, H, N).

Recombinant *Plasmodium* HGXPRT and human HGPRT were expressed and purified as previously described.[20] The recombinant human HGPRT has three of the four cysteine residues (C22, C105, C205) replaced by alanine to stabilize the enzyme. The kinetic and structural properties of this enzyme are identical to wild-type human HGPRT.[19,20]

5.1. Determination of K_i values

The K_i values were determined using a spectrophotometric assay at 25 °C, 0.1 M Tris–HCl, 10 mM $MgCl_2$, pH 7.4[19] The K_i values are $K_{i(app)}$ as they were measured at a single concentration of the second substrate. They approximate K_i values since the concentration of the second substrate (guanine) was saturating: 60 μM, approximately 30-fold higher than the measured K_m. $K_{i(app)}$ was calculated using the equations

$$v = V_{max}[S]_o/[S]_o + K_{m(app)} \text{ and } K_{m(app)} = K_m(1 + [I]/K_{i(app)}).$$

5.2. Reduction of substituted triethyl phosphonoacetates—general procedure[16]

Substituted triethyl phosphonoacetate (40 mmol) was cooled to –20 °C and solution of borane in THF (1.0 M, 60 ml) was added under argon atmosphere. The reaction mixture was stirred at room temperature for two days. Methanol (20 ml) was added at –20 °C and the solution was concentrated after the evolution of hydrogen stopped. Chloroform (250 ml) was added and the mixture was washed with saturated NaCl solution. The chloroform solution was dried over anhydrous magnesium sulfate and then evaporated to give the crude product that was purified by chromatography on silica gel (hexane–$CHCl_3$–MeOH).

5.2.1. Diethyl 1-hydroxypropan-2-yl-phosphonate (2a)

Starting from triethyl 2-phosphonopropionate, yield 90%, colorless oil. ^{1}H NMR (DMSO-d_6): 3.97 m, 4H (P–OCH_2); 3.68 dt, 1H, J(OH,1) = 4.3, J(OH,P) = 10.33 (OH); 3.44 q, 1H, J = 6.7 (H-1a); 3.30 td, 1H, J = 8.5 and 10.6 (H-1b); 1.95 m, 1H (H-2); 1.21 t, 6H, $J(CH_3,CH_2)$ = 7.1 (CH_3); 1.08 dd, 3H, J(3,2) = 7.14, J(3,P) = 17.1 (H-3). ^{13}C NMR (DMSO-d_6): 60.72 (C-1); 60.63 d, 2C, J(P,C) = 3.6 (P–OC); 33.76 d, J(P,C) = 135.2 (C-2); 16.17 d, 2C, J(P,C) = 5.5 (CH_3); 10.82 d, J(P,C) = 5.0 (C-3). MS (ESI): m/z = 197 $[M+H]^+$.

5.2.2. Diethyl 1-hydroxybutan-2-yl-phosphonate (2b)

Starting from triethyl 2-phosphonobutyrate, yield 79%, colorless oil. ^{1}H NMR (DMSO-d_6): 4.68 t, 1H, J(OH,1) = 5.0 (OH); 3.98 m, 4H (P–OCH$_2$); 3.66 m, 1H, (H-1a); 3.45 m, 1H (H-1b); 1.79 m, 1H (H-2); 1.61 m, 2H (H-3); 1.21 t, 6H, J(CH$_3$,CH$_2$) = 7.0 (CH$_3$); 0.96 t, 3H, J(4,3) = 7.5 (H-4). ^{13}C NMR (DMSO-d_6): 60.61 d, 2C, J(P,C) = 6.4 (P–OC); 58.58 (C-1); 40.50 d, J(P,C) = 129.8 (C-2); 18.54 d, J(P,C) = 3.7 (C-3), 16.24 d, 2C, J(P,C) = 5.6 (CH$_3$); 11.96 d, J(P,C) = 8.0 (C-4). MS (ESI): m/z = 211 [M+H]$^+$.

5.2.3. Diethyl 1-hydroxy-3-phenylpropan-2-yl-phosphonate (2c)

Starting from triethyl 3-phenyl-2-phosphonopropionate,[15] yield 87%, colorless oil. ^{1}H NMR (DMSO-d_6): 7.24 m, 5H (Ar); 4.79 t, 1H, J(OH,1) = 4.8 (OH); 3.93 m, 4H (P–OCH$_2$); 3.51 m, 2H (H-1); 2.85 m, 2H (H-3); 2,16 m, 1H (H-2); 1.6 dt, 6H, J = 7.0 and 5.9 (CH$_3$). ^{13}C NMR (DMSO-d_6): 139.55 d, J(P,C) = 10.5 (Ar); 128.94, 2C (Ar); 128.00, 2C (Ar); 125.90 (Ar); 60.69 d, 2C, J(P,C) = 6.4 (P–OC); 58.26 d, J(P,C) = 1.9 (C-1); 40.90 d, J(P,C) = 134.5 (C-2); 31.10 d, J(P,C) = 2.8 (C-3); 16.12 d, J(P,C) = 3.0 (CH$_3$). MS (ESI): m/z = 273 [M+H]$^+$.

5.3. Diethyl 2-hydroxyalkylphosphonates (22)

Diethyl 2-hydroxyalkylphosphonates **22** were prepared according the known procedure.[18]

5.3.1. Diethyl 2-hydroxypropylphosphonate (22a)

^{1}H NMR (DMSO-d_6): 4.74 d, 1H, J = 1.2 (OH); 3.97 m, 5H (H-2 and POCH$_2$); 1.87 m, 2H (H-1); 1.22 t, 6H, J(CH$_3$,CH$_2$) = 7.1 (CH$_3$); 1.16 dd, 3H, J = 6.2 and 0.6 (H-3). ^{13}C NMR (DMSO-d_6): 61.76 d, J(P,C) = 0.9 (C-2); 60.77 d and 60.56 d, J(P,C) = 6.1 (POCH$_2$); 35.12 d, J(P,C) = 133.3 (C-1); 24.41 d, J(P,C) = 8.8 (C-3); 16.17 d, J(P,C) = 5.8 (CH$_3$).

5.3.2. Diethyl 2-hydroxybutylphosphonate (22b)

^{1}H NMR (DMSO-d_6): 4.67 d, 1H, J = 5.3 (OH); 3.97 m, 4H (POCH$_2$); 3.69 m, 1H (H-2); 1.87 ddd, 2H, J = 6.4 and 2.1, J(1,P) = 17.9 (H-1); 1.54 dqd, 1H, J = 7.4 and 4.3, J_g = 14.9 (H-3a); 1.36 m, 1H (H-3b); 1.22 t, 6H, J(CH$_3$,CH$_2$) = 7.1 (CH$_3$); 0.85 t, 3H, J(4,3) = 7.4 (H-4). ^{13}C NMR (DMSO-d_6): 66.53 d, J(P,C) = 2.5 (C-2); 60.78 d, and 60.51, J(P,C) = 6.2 (POCH$_2$); 33.01 d, J(P,C) = 135.1 (C-1); 30.51 d, J(P,C) = 10.8 (C-3); 16.16 d, J(P,C) = 5.9 (CH$_3$); 9.59 (C-4). MS (ESI): m/z = 233 [M+Na]$^+$.

5.3.3. Diethyl 2-hydroxy-3-phenylpropylphosphonate (22c)

^{1}H NMR (DMSO-d_6): 7.28 m, 3H and 7.20 m, 2H (Ar); 4.90 d, 1H (OH); 3.97 m, 5H (POCH$_2$ and H-2); 2.82 dd, 1H, J(3a,2) = 5.2, J_g = 13.4 (H-3a); 2.70 ddd, 1H, J(3b,2) = 7.2 and 1.2, J_g = 13.5 (H-3b); 1.86 dd, 2H, J(1,2) = 6.4, J(1,P) = 17.8 (H-1); 1.21 dt, 6H, J(CH$_3$,CH$_2$) = 7.0, J(CH$_3$,P) = 1.1 (CH$_3$). ^{13}C NMR (DMSO-d_6): 138.77 (Ar); 129.36, 2C (Ar); 127.95, 2C (Ar); 125.86 (Ar); 66.73 d, J(P,C) = 2.8 (C-2); 60.86 d and 60.57 d, J(P,C) = 6.1 (POCH$_2$); 43.94 d, J(P,C) = 11.7 (C-3); 32.84 d, J(P,C) = 135.0 (C-1); 16.15 d, J(P,C) = 6.0 (CH$_3$). MS (ESI): m/z = 273 [M+H]$^+$.

5.4. Synthesis of diethyl (2-hydroxyethoxy)alkylphosphonates 9 and 24—general procedure

To the mixture of NaH (1.7 g, 60% disp. in mineral oil), dry THF (60 ml) and dry Et$_3$N (8 ml) cooled to −40 °C under argon atmosphere hydroxy derivative (**2a**–**2c** or **22a**–**22c**, 40 mmol) was added. After 30 min of vigorous stirring ethyl bromoacetate (5.5 ml, 50 mmol) was added and the temperature of the reaction mixture was allowed to rise slowly to room temperature. After 5 h of stirring diethyl ether (250 ml) was added and the mixture was washed with saturated NaCl solution. The organic layer was dried over anhydrous magnesium sulfate and then evaporated to give the crude intermediate that was used in following step.

5.4.1. Ethyl 2-(2-(diethoxyphosphoryl)propoxy)acetate (8a)

Starting from diethyl 1-hydroxypropan-2-yl-phosphonate (**2a**). ^{1}H NMR (DMSO-d_6): 4.12 s, 2H (CH$_2$COOEt); 4.12 q, 2H, J = 7.1 (COOCH$_2$); 3.99 m, 4H (P–OCH$_2$); 3.69 ddd, 1H, J(1a,2) = 4.3, J_g = 9.5, J(1a,P) = 10.2 (H-1a); 3.41 dd, 1H, J = 9.1 and 17.4 (H-1b); 2.20 m, 1H (H-2); 1.22 t, 6H, J(CH$_3$,CH$_2$P) = 7.0 (CH$_3$); 1.20 t, 3H, J(CH$_3$,CH$_2$) = 7.2 (CH$_3$); 1.11 dd, 3H, J(3,2) = 7.1, J(3,P) = 18.0 (H-3). ^{13}C NMR (DMSO-d_6): 169.80 (CO); 70.16 (COCH$_2$O); 67.34 (C-2); 60.91 d, 2C, J(P,C) = 6.3 (P–OC); 59.95 (C-1); 31.16 d, J(P,C) = 137.9 (C-2); 16.12 d, 2C, J(P,C) = 5.6 (CH$_3$); 13.88 (CH$_3$); 11.11 d, J(P,C) = 5.2 (C-3). MS (ESI): m/z = 283 [M+H]$^+$.

5.4.2. Ethyl 2-(2-(diethoxyphosphoryl)butoxy)acetate (8b)

Starting from diethyl 1-hydroxybutan-2-yl-phosphonate (**2b**). ^{1}H NMR (DMSO-d_6): 4.12 q, 2H, J = 7.2 (COOCH$_2$); 4.11 s, 2H (CH$_2$COOEt); 3.98 m, 4H (P–OCH$_2$); 3.69 ddd, 1H, J(1a,2) = 4.2, J_g = 9.5, J(1a,P) = 15.5 (H-1a); 3.55 dt, 1H, J= 9.3and 7.5 (H-1b); 2.00 m, 1H (H-2); 1.63 m, 2H (H-3); 1.22 q, 6H, J(CH$_3$,CH$_2$P) = 7.5 (CH$_3$); 1.21 t, 3H, J(CH$_3$,CH$_2$) = 7.0 (CH$_3$); 0.98 t, 3H, J(4,3) = 7.5 (H-4). ^{13}C NMR (DMSO-d_6): 169.89 (CO); 68.34 (COCH$_2$O); 67.54 (C-2); 60.85 d, 2C, J(P,C) = 6.4 (P–OC); 60.05 (C-1); 37.96 d, J(P,C) = 135.7 (C-2); 19.02 d, J(P,C) = 3.8 (C-3); 16.21 d, 2C, J(P,C) = 5.6 (CH$_3$); 13.97 (CH$_3$); 11.75 d, J(P,C) = 8.5 (C-4). MS (ESI): m/z = 297 [M+H]$^+$.

5.4.3. Ethyl 2-(2-(diethoxyphosphoryl)-3-phenylpropoxy)acetate (8c)

Starting from diethyl 1-hydroxy-3-phenylpropan-2-yl-phosphonate (**2c**). ^{1}H NMR (DMSO-d_6): 7.25 m, 5H (Ar); 4.09 m, 4H, (COOCH$_2$ and CH$_2$COOEt); 3.96 m, 4H (P–OCH$_2$); 3.56 m, 2H (H-1); 2.87 m, 2H (H-3); 2.37 m, 1H (H-2); 1.17 m, 9H, (CH$_3$). MS (ESI): m/z = 359 [M+H]$^+$.

5.4.4. Ethyl 2-[1-(diethoxyphosphoryl)propan-2-yloxy]acetate (23a)

Starting from diethyl 2-hydroxypropylphosphonate (**22a**). ^{1}H NMR (DMSO-d_6): 4.11 q, 2H, J = 7.1 (COOCH$_2$); 4.11 d, 2H (CH$_2$COOEt); 3.97 m, 4H (P–OCH$_2$); 3.74 m, 1H (H-2); 2.17 ddd, 1H, J = 19.7, 15.2 and 4.85 (H-1a); 1.89 m, 1H (H-1b); 1.21 m, 12H (H-3, CH$_3$). ^{13}C NMR (DMSO-d_6): 160.02 (CO); 71.18 (C-2); 65.30 (COCH$_2$O); 60.89 d and 60.74, J(P,C) = 6.1 (P–OC); 31.99 d, J(P,C) = 134.5 (C-2); 20.68 d, J(P,C) = 6.2 (C-3); 16.11 d, 2C, J(P,C) = 5.7 (CH$_3$); 13.96 (CH$_3$). MS (ESI): m/z = 283 [M+H]$^+$.

5.4.5. Ethyl 2-[1-(diethoxyphosphoryl)butan-2-yloxy]acetate (23b)

Starting from diethyl 2-hydroxybutylphosphonate (**22b**). ^{1}H NMR (DMSO-d_6): 4.11 dd 2H, J = 14.3 and 7.1 (COOCH$_2$); 4.13 s, 2H (CH$_2$COOEt); 3.98 m, 4H (P–OCH$_2$); 3.59 m, 1H (H-2); 2.12 ddd, 1H, J = 19.4, 15.4 and 5.3 (H-1a); 1.92 ddd, 1H, J = 18.0, 15.4 and 7.3 (H-1b); 1.68 ddd, 1H, J = 14.1, 7.3 and 4.6 (H-3a); 1.53 dt, 1H, J = 14.1 and 7.1 (H-3b); 1.21 t, 6H, J = 7.2, (CH$_3$); 1.19 t, 6H, J = 7.2, (CH$_3$); 0.88 t, 3H, J = 7.4, (H-4). ^{13}C NMR (DMSO-d_6): 170.19 (CO); 75.99 (C-2); 61.07 d and 60.93, J(P,C) = 6.1 (P–OC); 60.18 (COCH$_2$O); 29.48 d, J(P,C) = 134.8 (C-2); 27.27 d, J(P,C) = 4.8 (C-3); 16.28 d, 2C, J(P,C) = 5.9 (CH$_3$); 14.14 (CH$_3$); 9.05 (C-4). MS (ESI): m/z = 297 [M+H]$^+$.

5.4.6. Ethyl 2-[1-(diethoxyphosphoryl)-3-phenylpropan-2-yloxy]acetate (23c)

Starting from diethyl 2-hydroxy3-phenylpropylphosphonate (**22c**). ^{1}H NMR (DMSO-d_6): 7.29 m, 5H (Ar); 4.12 m, 4H (COOCH$_2$

and CH_2COOEt); 3.96 m, 4H ($POCH_2$); 3.83 m, 1H (H-2); 2.87 d, 2H, J(5,3) = 5.6 (H-3); 2.08 m, 2H (H-1); 1.22 dt, 6H, $J(CH_3,CH_2)$ = 7.1, $J(CH_3,P)$ = 2.5 (CH_3). MS (ESI): m/z = 359 $[M+H]^+$.

Crude intermediate (**8a–8c** or **23a–23c**) was cooled to –20 °C and a solution of borane in THF (1.0 M, 50 ml) was added under argon atmosphere. The reaction mixture was stirred at room temperature for three days. Methanol (20 ml) was added at –20 °C and the solution was concentrated after the evolution of hydrogen had stopped. The residue was purified by chromatography on silica gel (hexane–$CHCl_3$–MeOH).

5.4.7. Diethyl 1-(2-hydroxyethoxy)propan-2-yl-phosphonate (9a)

Starting from intermediate **8a**, yield 58% in two steps, colorless oil. ^{1}H NMR (DMSO-d_6): 4.59 br s, 1H (OH); 3.99 m, 4H (P–OCH_2); 3.60 ddd, 1H, J = 4.6, 9.7 and 10.73 (H-3a); 3.48 dd, 2H J = 4.8 and 4.5 and (H-2); 3.39 m, 3H (H-3b and H-1); 2.16 m, 1H (H-4); 1.22 t, 6H, $J(CH_3,CH_2)$ = 7.1 (CH_3); 1.09 dd, 3H, J(5,4) = 7.13, J(5,P) = 18.0 (H-5). ^{13}C NMR (DMSO-d_6): 72.05 and 69.95 (C-3 and C-2); 60.97 d, 2C, J(P,C) = 2.4 (P–OC); 60.04 (C-1); 31.26 d, J(P,C) = 137.6 (C-4); 16.23 d, 2C, J(P,C) = 5.6 (CH_3); 11.25 d, J(P,C) = 5.1 (C-5). MS (ESI): m/z = 241 $[M+H]^+$.

5.4.8. Diethyl 1-(2-hydroxyethoxy)butan-2-yl-phosphonate (9b)

Starting from intermediate **8b**, yield 64% in two steps, colorless oil. ^{1}H NMR (DMSO-d_6): 4.58 t, 1H, J = 5.4, (OH); 3.98 m, 4H (P–OCH_2); 3.61 ddd, 1H, J = 4.5, 9.8 and 15.8 (H-3a); 3.50 m, 1H (H-3b); 3.49 t, 2H, J = 5.5 (H-2); 3.40 dt, 2H, J = 5.0 and 10.2 (H-1); 1.97 m, 1H (H-4); 1.59 m, 2H (H-5); 1.22 t, 6H, $J(CH_3,CH_2)$ = 7.1 (CH_3); 0.96 t, 3H, J(5,4) = 7.5 (H-6). ^{13}C NMR (DMSO-d_6): 72.12 and 67.97 (C-3 and C-2); 60.84 d and 60.86 d, J(P,C) = 6.5 (P–OC); 60.01 (C-1); 38.00 d, J(P,C) = 135.4 (C-4); 19.10 d, J(P,C) = 3.9 (C-5); 16.23 d, 2C, J(P,C) = 4.9 (CH_3); 11.88 d, J(P,C) = 8.5 (C-6). MS (ESI): m/z = 255 $[M+H]^+$.

5.4.9. Diethyl 1-(2-hydroxyethoxy)-3-phenylpropan-2-yl-phosphonate (9c)

Starting from intermediate **8c**, yield 77% in two steps, colorless oil. ^{1}H NMR (DMSO-d_6): 7.24 m, 5H (Ar); 4.58 t, 1H, J = 5.5, (OH); 3.95 m, 4H (P–OCH_2); 3.47 m, 4H (H-2 and H-3); 3.35 m, 2H (H-1); 2.85 m, 2H (H-5); 2.32 m, 1H (H-4); 1.18 dd, 6H, J = 7.2 and 14.7 (CH_3). ^{13}C NMR (DMSO-d_6): 139.09 d, J(P,C) = 11.4 (Ar); 128.98, 2C (Ar); 128.05, 2C (Ar); 126.01 (Ar); 72.15 and 67.31 (C-3 and C-2); 60.98 d and 60.83, J(P,C) = 6.3 (P–OC); 58.93 (C-1); 37.89 (C-4); 31.45 d, J(P,C) = 2.7 (C-5); 16.13 d, 2C, J(P,C) = 3.6 (CH_3). MS (ESI): m/z = 317 $[M+H]^+$.

5.4.10. Diethyl 2-(2-hydroxyethoxy)propylphosphonate (24a)

Starting from intermediate **23a**, yield 69% in two steps, colorless oil. ^{1}H NMR (DMSO-d_6): 3.98 m, 5H (H-3 and $POCH_2$); 3.70 m, 1H (H-2); 3.37 m, 2H (H-1); 2.06 ddd, 1H, J = 18.5, 15.3 and 6.3 (H-4a); 1.88 ddd, 1H, J = 17.7, 15.3 and 6.6 (H-4b); 1.22 t, 6H, $J(CH_3,CH_2)$ = 7.3 (CH_3); 1.18 d, 3H, J(5,3) = 6.0 (H-5). ^{13}C NMR (DMSO-d_6): 70.39 (C-3); 69.49 (C-1); 60.92 d and 60.69 d, J(P,C) = 6.2 ($POCH_2$); 60.24 (C-2); 32.41 d, J(P,C) = 135.4 (C-4); 20.77 d, J(P,C) = 9.5 (C-5); 16.15 d, J(P,C) = 6.0 (CH_3). MS (ESI): m/z = 241 $[M+H]^+$.

5.4.11. Diethyl 2-(2-hydroxyethoxy)butylphosphonate (24b)

Starting from intermediate **23b**, yield 73% in two steps, colorless oil. ^{1}H NMR (DMSO-d_6): 4.61 t, 1H (OH); 3.99 m, 4H ($POCH_2$); 3.44 m, 5H (H-1, H-2 and H-3); 3.37 m, 2H (H-1); 1.95 m, 2H (H-4); 1.55 m, 2H (H-5); 1.22 t, 6H, $J(CH_3,CH_2)$ = 7.2 (CH_3); 0.85 t, 3H, J(5,3) = 6.2 (H-5). ^{13}C NMR (DMSO-d_6): 70.39 (C-3); 69.49 (C-1); 60.91 d and 60.68 d, J(P,C) = 6.2 ($POCH_2$); 60.24 (C-2); 32.41 d, J(P,C) = 135.4 (C-4); 20.77 d, J(P,C) = 9.5 (C-5); 16.15 d, J(P,C) = 6.0 (CH_3). MS (ESI): m/z = 255 $[M+H]^+$.

5.4.12. Diethyl 2-(2-hydroxyethoxy)-3-phenylpropylphosphonate (24c)

Starting from intermediate **23c**, yield 81% in two steps, colorless oil. ^{1}H NMR (DMSO-d_6): 7.26 m, 5H (Ar); 4.61 br s, 1H (OH); 3.95 m, 4H ($POCH_2$); 3.81 m, 1H (H-3); 3.44 m, 4H (H-2 and H-1); 2.85 d, 2H, J(5,3) = 5.7 (H-5); 1.90 m, 2H (H-4); 1.20 dt, 6H, $J(CH_3,CH_2)$ = 7.0, $J(CH_3,P)$ = 2.6 (CH_3). ^{13}C NMR (DMSO-d_6): 137.96 (Ar); 129.48, 2C (Ar); 128.07, 2C (Ar); 126.08 (Ar); 75.46 d, J(P,C) = 3.1 (C-3); 70.48 (C-1); 60.85 dd, J(P,C) = 6.2 and 27.8 ($POCH_2$); 60.15 (C-2); 40.25 d, J(P,C) = 12.0 (C-5); 29.94 d, J(P,C) = 137.3 (C-4); 16.12 dd, J(P,C) = 2.0 and 5.9 (CH_3). MS (ESI): m/z = 317 $[M+H]^+$.

5.5. Synthesis of diethyl *O*-toluensulfonyl-1-(2-hydroxyethoxy)alkan-2-yl-phosphonates 10—general procedure

Solution of diethyl 1-(2-hydroxyethoxy)-alkan-2-yl-phosphonate (**9a–9c**, 4.7 mmol) in dichloromethane (20 ml) was cooled to –10 °C and then dimethylaminopyridine (50 mg), dry triethylamine (1 ml) and toluensulfonylchloride (1.16 g, 6.1 mmol) were added. The reaction mixture was stirred at room temperature overnight. Water with ice and Et_2O were added, the organic layer was separated, washed with brine and dried over anhydrous $MgSO_4$. After filtration, solvent was evaporated and the residue was purified by column chromatography on silica gel (hexane–$CHCl_3$–MeOH) to afford the product as colorless oil.

5.5.1. Diethyl *O*-toluensulfonyl-1-(2-hydroxyethoxy)propan-2-yl-phosphonate (10a)

Starting from hydroxy derivative **9a**, yield 66%. ^{1}H NMR (DMSO-d_6): 7.78 d, 2H, J = 8.2 and 7.48 d, 2H, J = 8.1 (Ar); 4.11 dd, 2H, J(1,2) = 4.3 (H-1); 3.96 m, 4H (P–OCH_2); 3.56 m, 3H (H-2 and H-3a); 3.28 m, 1H (H-3b); 2.42 s, 3H (CH_3Ar); 2.07 m, 1H (H-4); 1.20 t, 6H, J = 7.1 (CH_3); 1.01 dd, 3H, J = 7.1 and 7.9 (H-6). MS (ESI): m/z = 395 $[M+H]^+$.

5.5.2. Diethyl *O*-toluensulfonyl-1-(2-hydroxyethoxy)butan-2-yl-phosphonate (10b)

Starting from hydroxy derivative **9b**, yield 50%. ^{1}H NMR (DMSO-d_6): 7.78 d, 2H, and 7.48 d, 2H (Ar); 4.11 dd, 2H, J(1,2) = 4.8 and 3.8 (H-1); 3.95 qd, 4H, J = 7.1 and 14.3 (P–OCH_2); 3.56 m, 3H (H-2 and H-3a); 3.42 dt, 1H, J = 7.13 and 10.0 (H-3b); 2.42 s, 3H (CH_3Ar); 1.89 m, 1H (H-4); 1.52 m, 2H (H-5); 1.19 dt, 6H, J = 7.1 and 0.8 (CH_3); 0.91 t, 3H, J(5,4) = 7.5 (H-6). ^{13}C NMR (DMSO-d_6): 144.81, 132.28, 130.04, 2C and 127.49, 2C (Ar); 69.80 (C-1); 68.05 and 67.74 (C-3 and C-2); 60.83 d, 2C, J(P,C) = 6.8 (P–OC); 37.86 d, J(P,C) = 141.1 (C-4); 20.99 (CH_3Ar); 18.98 d, J(P,C) = 3.0 (C-5); 16.18 d, 2C, J(P,C) = 5.5 (CH_3); 11.81 d, J(P,C) = 8.6 (C-6). MS (ESI): m/z = 409 $[M+H]^+$.

5.5.3. Diethyl *O*-toluensulfonyl-1-(2-hydroxyethoxy)-3-phenylpropan-2-yl-phosphonate (10c)

Starting from hydroxy derivative **9c**, yield 60%. ^{1}H NMR (DMSO-d_6): 7.78 d, 2H, J = 8.2 and 7.48 d, 2H, J = 8.1 (Ar); 7.22 m, 5H (Ar); 4.09 t, 2H, J(1,2) = 4.2 (H-1); 3.92 m, 4H (P–OCH_2); 3.50 m, 2H (H-2); 3.42 m, 1H (H-3a); 3.31 m, 1H (H-3b); 2.84 m, 1H (H-5′a); 2.72 m, 1H (H-5′b); 2.40 s, 3H (CH_3Ar); 2.28 m, 1H (H-4); 1.15 dt, 6H, J = 7.0 and 3.4 (CH_3). ^{13}C NMR (DMSO-d_6): 144.82, 138.93, J(P,C) = 11.4; 132.25, 128.93, 2C; 128.05, 2C; 127.53, 2C; 127.48 2C and 126.03 (Ar); 69.71 (C-1); 67.82 and 67.41 (C-3 and C-2); 60.91 d, 2C, J(P,C) = 6.7 (P–OC); 38.40 d, J(P,C) = 136.2 (C-4);

31.33 d, J(P,C) = 2.4 (C-5); 20.98 (CH_3Ar); 16.12 d, 2C, J(P,C) = 6.3 (CH_3). MS (ESI): m/z = 4717 $[M+H]^+$.

5.6. Alkylation of adenine—general procedure

A mixture of adenine (140 mg, 1 mmol), DMF (10 ml) and Cs_2CO_3 (200 mg, 0.5 mmol) was heated at 100 °C for 1 h. Tosyl derivative **10a**–**10c** in DMF (2 ml) was added and the reaction mixture was heated for 24 h. Solvent was evaporated, the residue was codistilled with toluene and ethanol. Chromatography on preparative TLC (10% MeOH–$CHCl_3$) afforded pure product as yellowish foam.

5.6.1. Diethyl 9-[2-(2-phosphonopropoxy)ethyl]adenine (11a)

Starting from tosyl derivative **10a**, yield 70%. ^{1}H NMR (DMSO-d_6): 8.13 s, 1H and 8.10 s, 1H (H-2, H-8); 7.19 br s, 2H (NH_2); 4.30 t, 2H, J(1′,2′) = 5.3 (H-1′); 3.93 m, 4H (P–OCH_2); 3.77 m, 2H (H-2′); 3.57 ddd, 1H, J(3′a,4′) = 4.6, J_g = 9.6, J(3′a,P) = 11.79 (H-3′a); 3.36 m, 1H (H-3′b); 2.10 m, 1H (H-4′); 1.17 t, 6H, J(CH_3,CH_2) = 7.0 (CH_3); 0.96 dd, 3H, J(5′,4′) = 7.13, J(5′,P) = 17.9 (H-5′). ^{13}C NMR (DMSO-d_6): 155.83 (C-6); 152.23 (C-2); 149.43 (C-4); 141.06 (C-8); 118.48 (C-5); 69.84 (C-3′); 68.07 (C-2′); 60.96 d, 2C, J(P,C) = 6.4 (P–OC); 42.58 (C-1′); 31.10 d, J(P,C) = 137.8 (C-4′); 16.22 d, 2C, J(P,C) = 5.5 (CH_3); 11.06 d, J(P,C) = 5.2 (C-5′). MS (ESI): m/z = 358 $[M+H]^+$.

5.6.2. Diethyl 9-[2-(2-phosphonobutoxy)ethyl]adenine (11b)

Starting from tosyl derivative **10b**, yield 65%. ^{1}H NMR (DMSO-d_6): 8.13 s, 1H and 8.10 s, 1H (H-2, H-8); 7.20 br s, 2H (NH_2); 4.30 t, 2H, J(1′,2′) = 5.2 (H-1′); 3.90 m, 4H (P–OCH_2); 3.77 m, 2H (H-2′); 3.54 ddd, 1H, J(3′a,4′) = 4.5, J_g = 9.8, J(3′a,P) = 17.2 (H-3′a); 3.51 td, 1H, J(3′b,4′) = 6.8, J_g = J(3′b,P) = 9.8 (H-3′b); 1.90 m, 1H (H-4′); 1.45 m, 2H (H-5′); 1.16 t, 6H, J(CH_3,CH_2) = 7.1 (CH_3); 0.81 t, 3H, J(6′,5′) = 7.5 (H-6′). ^{13}C NMR (DMSO-d_6): 155.77 (C-6); 152.14 (C-2); 149.41 (C-4); 141.06 (C-8); 118.50 (C-5); 68.22 (C-3′); 67.89 (C-2′); 60.77 d, 2C, J(P,C) = 6.6 (P–OC); 42.62 (C-1′); 37.85 d, J(P,C) = 138.0 (C-4′); 18.90 d, J(P,C) = 4.0 (C-5′); 16.17 d, 2C, J(P,C) = 6.0 (CH_3); 11.79 d, J(P,C) = 9.8 (C-6′). MS (ESI): m/z = 372 $[M+H]^+$.

5.6.3. Diethyl 9-[2-(3-phenyl 2-phosphonopropoxy)ethyl]adenine (11c)

Starting from tosyl derivative **10c**, yield 63%. ^{1}H NMR (DMSO-d_6): 8.14 s, 1H and 8.09 s, 1H (H-2, H-8); 7.15br s, 2H (NH_2); 7.17 m, 3H and 6.95 m, 2H (Ar); 4.29 t, 2H, J(1′,2′) = 5.0 (H-1′); 3.89 m, 4H (P–OCH_2); 3.70 dt, 1H and 3.65 td, 1H, J(2′,1′) = 4.9 and 5.4, J_g = 10.4 (H-2′); 3.48 dt, 1H, J(3′a,4′) = 5.0, J_g = 10.4 (H-3′a); 3.26 dd, 1H, J(3′a,4′) = 5.0, J_g = 9.5 (H-3′b); 2.79 dt, 1H, J(5′a,4′) = 5.2, J(5′a,P) = 12.9, J_g= 13.1 (H-5′a); 2.63 dt, 1H, J(5′b,4′) = 9.4, J(5′a,P) = 13.0, J_g= 13.3 (H-5′b), 2.25 m, 1H (H-4′); 1.14 q, 6H, J= 7.1 (CH_3). ^{13}C NMR (DMSO-d_6): 155.77 (C-6); 152.15 (C-2); 149.32 (C-4); 141.03 (C-8); 138.66 d, J(P,C) = 11.3 (Ar); 128.70, 2C, 127.87, 2C and 125.88 (Ar); 118.47 (C-5); 68.17 (C-3′); 66.99 (C-2′); 60.78 d, 2C, J(P,C) = 6.0 (P–OC); 42.65 (C-1′); 38.33 d, J(P,C) = 132.4 (C-4′); 31.08 d, J(P,C) = 2.2 (C-5′); 16.03 d, 2C, J(P,C) = 6.1 (CH_3). MS (ESI): m/z = 434 $[M+H]^+$.

5.7. Synthesis of diethyl 9-[2-(phosphonoalkoxy)ethyl]-6-chloropurines 13 and 25 via Mitsunobu reaction—general procedure

To a solution of triphenylphosphine (4.72 g, 18 mmol) in dry THF (60 ml) cooled to –20 °C under argon atmosphere diisopropylazadicarboxylate (DIAD, 3.2 ml, 16.8 mmol) was added slowly. The mixture was stirred for 30 min and this preformed complex was added to the reaction mixture containing chloropurine (2.04 g, 13.2 mmol), dry THF (40 ml) and diethyl phosphonate **9a**–**9c** or **24a**–**24c**, (6 mmol) at –40 °C under argon. The resulting mixture was slowly warmed to room temperature and stirred overnight. Solvent was evaporated and the crude mixture was purified by chromatography on silica gel (MeOH–$CHCl_3$). Pure product was obtained as yellowish foam.

5.7.1. Diethyl 9-[2-(2-phosphonopropoxy)ethyl]-6-chloropurine (13a)

Starting from hydroxy derivative **9a**, yield 39%. ^{1}H NMR (DMSO-d_6): 8.79 s, 1H and 8.69 s, 1H (H-2, H-8); 4.47 t, 2H, J(1′,2′) = 5.25 (H-1′); 3.91 m, 4H (P–OCH_2); 3.82 m, 2H (H-2′); 3.57 ddd, 1H, J(3′a,4′) = 4.6, J_g = 9.6, J(3′a,P) = 12.1 (H-3′a); 3.37 td, 1H, J(3′b,4′) = 8.3, J_g = J(3′b,P) = 9.7 (H-3′b); 2.09 m, 1H (H-4′); 1.16 t, 6H, J(CH_3,CH_2) = 7.1 (CH_3); 0.93 dd, 3H, J(5′,4′) = 7.2, J(5′,P) = 17.9 (H-5′). ^{13}C NMR (DMSO-d_6): 151.49 (C-4); 150.95 (C-2); 148.39 (C-6); 147.33 (C-8); 130.17 (C-5); 69.45 (C-3′); 67.27 (C-2′); 60.49 d, 2C, J(P,C) = 6.2 (P–OC); 43.03 (C-1′); 30.63 d, J(P,C) = 138.3 (C-4′); 15.75 d, 2C, J(P,C) = 5.6 (CH_3); 10.58 d, J(P,C) = 5.2 (C-5′). MS (ESI): m/z = 377–379 $[M+H]^+$.

5.7.2. Diethyl 9-[2-(2-phosphonobutoxy)ethyl]-6-chloropurine (13b)

Starting from the hydroxy derivative **9b**, yield 34%. ^{1}H NMR (DMSO-d_6): 8.79 s, 1H and 8.69 s, 1H (H-2, H-8); 4.48 t, 2H, J(1′,2′) = 5.10 (H-1′); 3.89 m, 4H (P–OCH_2); 3.83 m, 2H (H-2′); 3.53 m, 2H (H-3′); 1.87 m, 1H (H-4′); 1.40 m, 2H (H-5′); 0.76 t, 3H, J(6′,5′) = 7.5 (H-6′). ^{13}C NMR (DMSO-d_6): 151.92 (C-4); 151.35 (C-2); 148.80 (C-6); 147.74 (C-8); 130.60 (C-5); 67.90 (C-3′); 67.83 (C-2′); 60.75 d, 2C, J(P,C) = 6.3 (P–OC); 43.49 (C-1′); 37.75 d, J(P,C) = 135.8 (C-4′); 18.84 d, J(P,C) = 3.7 (C-5′); 16.11 d, 2C, J(P,C) = 5.9 (CH_3); 11.60 d, J(P,C) = 9.0 (C-6′). MS (ESI): m/z = 391–393 $[M+H]^+$.

5.7.3. Diethyl 9-[2-(3-phenyl-2-phosphonopropoxy)ethyl]-6-chloropurine (13c)

Starting from the hydroxy derivative **9c**, yield 68%. ^{1}H NMR (DMSO-d_6): 8.79 s, 1H and 8.69 s, 1H (H-2 and H-8); 7.15 m, 3H and 6.95 m, 2H (Ar); 4.47 t, 2H, J(1′,2′) = 5.12 (H-1′); 3.88 m, 4H (P–OCH_2); 3.74 m, 2H (H-2′); 3.49 m, 1H and 3.32 m, 1H (H-3′); 2.77 ddd, 1H, J(5′a,4′) = 9.2, J_g = 12.5, J(5′a,P) = 13.6 (H-5′a); 2.59 ddd, 1H, J(5′b,4′) = 9.1, J(5′b,P) = 12.5, J_g = 13.6 (H-5′b); 2.27 m, 1H (H-4′); 1.12 q, 6H, J(CH_3,CH_2) = 7.0 (CH_3). ^{13}C NMR (DMSO-d_6): 157.89 (C-4); 151.37 (C-2); 148.85 (C-6); 147.80 (C-8); 138.74 d, J(P,C) = 12.0 (Ar); 130.65 (C-5); 128.69, 2C (Ar); 127.94, 2C (Ar); 126.00 (Ar); 67.88 (C-2′); 67.21 (C-3′); 60.87 d, 2C, J(P,C) = 6.0 (P–OC); 43.56 (C-1′); 40.00 d, J(P,C) = 135.0 (C-4′); 31.17 (C-5′); 16.08 d, 2C, J(P,C) = 5.0 (CH_3). MS (ESI): m/z = 453–455 $[M+H]^+$.

5.7.4. Diethyl 9-[2-(1-phosphonopropan-2-yloxy)ethyl]-6-chloropurine (25a)

Starting from the hydroxy derivative **24a**, yield 35%. ^{1}H NMR (DMSO-d_6): 8.78 s, 1H and 8.69 s, 1H (H-2 and H-8); 4.30 t, 2H, J(1′,2′) = 5.1 (H-1′); 3.90 m, 4H ($POCH_2$); 3.83 m, 2H (H-2′); 3.67 m, 1H (H-3′); 1.98 ddd, 1H, J = 18.7, 15.3 and 5.9 (H-4′a); 1.83 ddd, 1H, J = 17.9, 15.3 and 6.9 (H-4′b); 1.16 dt, 6H, J = 7.0 and 4.6 (CH_3); 1.1 d, 3H, J = 6.1(H-5′).^{13}C NMR (DMSO-d_6): 152.22 (C-4); 151.66 (C-2); 149.12 (C-6); 148.12 (C-8); 130.95 (C-5); 71.07 (C-3′); 65.46 (C-2′); 61.06 d, 2C, J(P,C) = 6.3 ($POCH_2$); 44.26 (C-1′); 32.49 d, J(P,C) = 135.3 (C-4′), 20.87 d, J(P,C) = 8.5 (C-5′); 16.41 dd, J(P,C) = 1.1 and 5.8 (CH_3). MS (ESI): m/z = 377–379 $[M+H]^+$.

5.7.5. Diethyl 9-[2-(1-phosphonobutan-2-yloxy)ethyl]-6-chloropurine (25b)

Starting from the hydroxy derivative **24b**, yield 53%. ^{1}H NMR (DMSO-d_6): 8.78 s, 1H and 8.69 s, 1H (H-2 and H-8); 4.44 t, 2H,

$J(1',2')$ = 5.3 (H-1′); 3.91 m, 5H ($POCH_2$ and H-2′a); 3.80 m, 1H (H-2′b); 3.48 m, 1H (H-3′); 1.88 dddd, 2H, J = 42.7, 18.1, 15.4 and 6.3 (H-4′); 1.52 ddd, 1H, J = 14.1, 7.2 and 4.9 (H-5′a); 1.38 td, 1H, J = 14.2 and 7.1 (H-5′b);1.17 dt, 6H, J = 7.1 and 3.5 (CH_3); 0.61 t, 3H, J = 7.4 (H-6′). ^{13}C NMR (DMSO-d_6): 151.93 (C-4); 151.34 (C-2); 148.79 (C-6); 147.82 (C-8); 130.66 (C-5); 75.46 d, J(P,C) = 1.5 (C-3′); 65.56 (C-2′); 60.76 d, J(P,C) = 6.3 ($POCH_2$); 44.05 (C-1′); 29.46 d, J(P,C) = 136.4 (C-4′), 26.78 d, J(P,C) = 8.9 (C-5′); 16.11 dd, J(P,C) = 5.7 (CH_3); 8.52 (C-6′). MS (ESI): m/z = 391–393 $[M+H]^+$.

5.7.6. Diethyl 9-[2-(3-phenyl-1-phosphonopropan-2-yloxy)ethyl]-6-chloropurine (25c)

Starting from the hydroxy derivative **24c**, yield 51%. ^{1}H NMR (DMSO-d6): 8.74 s, 1H and 8.55 s, 1H (H-2 and H-8); 7.05 m, 3H and 6.93 m, 2H (Ar); 4.37 t, 2H, $J(1',2')$ = 5.0 (H-1′); 3.91 m, 5H (POCH2 and H-3′); 3.72 m, 2H (H-2′); 2.80 dd, 1H, $J(5'a,3')$ = 4.7, J_g = 13.7 (H-5′a); 2.67 dd, 1H, $J(5'b,3')$ = 7.2, J_g = 13.7 (H-5′b); 1.91 m, 2H (H-4′); 1.17 t, 6H, J(CH2CH3) = 7.0 (CH3). ^{13}C NMR (DMSO-d6): 151.78 (C-4); 151.19 (C-2); 148.75 (C-6); 147.63 (C-8); 137.69 (Ar); 130.69 (C-5); 129.01, 2C (Ar); 127.72, 2C (Ar); 125.82 (Ar); 75.65 d, J(P,C) = 2.1 (C-3′); 66.02 (C-2′); 60.81d, J(P,C) = 6.4 ($POCH_2$); 43.95 (C-1′); 40.32 d, J(P,C) = 10.0 (C-5′); 29.72 d, J(P,C) = 135.8 (C-4′), 16.09 d, J(P,C) = 5.8 (CH_3). MS (ESI): m/z = 453–455 $[M+H]^+$.

5.8. Synthesis of diethyl 9-[2-(phosphonoalkoxy)ethyl]-2-amino-6-chloropurines 14 and 26 via Mitsunobu reaction—general procedure

The procedure was identical as described above for 6-chloropurine, only after the stirring of reaction mixture overnight water (30 ml) was added and the mixture was heated at 80 °C for 20 h. Solvent was evaporated and the crude mixture was purified by chromatography on silica gel (MeOH–$CHCl_3$). Pure product was obtained as yellowish foam.

5.8.1. Diethyl 9-[2-(2-phosphonopropoxy)ethyl]-2-amino-6-chloropurine (14a)

Starting from the hydroxy derivative **9a**, yield 35%. ^{1}H NMR (DMSO-d_6): 8.10 s, 1H (H-8); 6.91 br s, 2H (NH_2); 4.21 t, 2H, $J(1',2')$ = 5.3 (H-1′); 3.93 m, 4H (P–OCH_2); 3.76 dt, 1H and 3.73 dt, 1H, $J(2',1')$ = 5.3, J_g = 10.7 (H-2′); 3.57 ddd, 1H, $J(3'a,4')$ = 4.6, J_g = 9.6, $J(3'a,P)$ = 11.9 (H-3′a); 3.36 td, 1H, $J(3'b,4')$ = 8.4, J_g = $J(3'b,P)$ = 9.6 (H-3′b); 2.105 m, 1H, $J(3',4')$ = 4.6 and 8.4, $J(4',5')$ = 7.2, $J(4',P)$ = 15.9 (H-4′); 1.18 t, 6H, $J(CH_3,CH_2)$ = 7.1 (CH_3); 0.98 dd, 3H, $J(5',4')$ = 7.2, $J(5',P)$ = 17.9 (H-5′). ^{13}C NMR (DMSO-d_6): 159.43 (C-2); 153.81 (C-4); 148.93 (C-6); 143.23 (C-8); 122.875 (C-5); 69.45 (C-3′); 67.49 (C-2′); 60.73 d, 2C, J(P,C) = 6.2 (P–OC); 42.47 (C-1′); 30.90 d, J(P,C) = 138.6 (C-4′); 15.97 d, 2C, J(P,C) = 5.6 (CH_3); 10.86 d, J(P,C) = 5.3 (C-5′). MS (ESI): m/z = 392–394 $[M+H]^+$.

5.8.2. Diethyl 9-[2-(2-phosphonobutoxy)ethyl]-2-amino-6-chloropurine (14b)

Starting from the hydroxy derivative **9b**, yield 48%. ^{1}H NMR (DMSO-d_6): 8.09 s, 1H (H-8); 6.90 br s, 2H (NH_2); 4.21 t, 2H, $J(1',2')$ = 5.2 (H-1′); 3.91 m, 4H (P–OCH_2); 3.77 m, 2H (H-2′); 3.53 m, 2H (H-3′); 1.54 m, 1H (H-4′); 1.45 m, 2H (H-5′); 1.16 t, 6H, $J(CH_3,CH_2)$ = 7.1 (CH_3); 0.82 t, 3H, $J(6',5')$ = 7.5 (H-6′). ^{13}C NMR (DMSO-d_6): 159.63 (C-2); 154.01 (C-4); 149.13 (C-6); 143.44 (C-8); 123.11 (C-5); 67.95 (C-3′); 67.82 (C-2′); 60.78 d, 2C, J(P,C) = 6.1 (P–OC); 42.73 (C-1′); 37.82 d, J(P,C) = 135.6 (C-4′); 18.90 d, J(P,C) = 3.8 (C-5′); 16.17 d, 2C, J(P,C) = 5.5 (CH_3); 11.68 d, J(P,C) = 8.9 (C-5′). MS (ESI): m/z = 406–408 $[M+H]^+$.

5.8.3. Diethyl 9-[2-(3-phenyl-2-phosphonopropoxy)ethyl]-2-amino-6-chloropurine (14c)

Starting from the hydroxy derivative **9c**, yield 55%. ^{1}H NMR (DMSO-d_6): 8.09 s, 1H (H-8); 7.16 m, 3H and 6.98 m, 2H (Ar); 6.92 br s, 2H (NH_2); 4.20 t, 2H, $J(1',2')$ = 5.11 (H-1′); 3.89 m, 4H ($POCH_2$); 3.65 m, 2H (H-2′); 3.48 m, 1H and 3.30 m, 1H (H-3′); 2.80 dt, 1H, $J(5'a,4')$ = 5.4, J_g and $J(5'a,P)$ = 13.0 and 13.3 (H-5′a); 2.64 dt, 1H, $J(5'b,4')$ = 9.1, $J(5'b,P)$ = 9.8, J_g and $J(5'b,P)$ = 13.1 and 13.4 (H-5′b); 2.27 m, 1H (H-4′), 1.14 td, 6H, J = 7.1 and 8.0 (CH_3). ^{13}C NMR (DMSO-d_6): 159.67 (C-2); 153.99 (C-4); 149.20 (C-6); 143.57 (C-8); 138.78 d, J(P,C) = 12.0 (Ar); 128.81, 2C (Ar); 127.99, 2C (Ar); 126.02 (Ar); 123.19 (C-5); 67.80 (C-2′); 67.19 d, J(P,C) = 1.9 (C-3′); 60.90 d, 2C, J(P,C) = 6.3 ($POCH_2$); 42.85 (C-1′); 38.06 d, J(P,C) = 135.3 (C-4′); 31.20 d, J(P,C) = 2.5 (C-5′); 16.125 t, J(P,C) = 5.6 (CH_3). MS (ESI): m/z = 468–470 $[M+H]^+$.

5.8.4. Diethyl 9-[2-(1-phosphonopropan-2-yloxy)ethyl]-2-amino-6-chloropurine (26a)

Starting from the hydroxy derivative **24a**, yield 63%. ^{1}H NMR (DMSO-d6): 8.11 s, 1H (H-8); 6.91 s, 2H (NH_2); 4.16 t, 2H, $J(1',2')$ = 5.2 (H-1′); 3.91 m, 4H ($POCH_2$); 3.74 m, 1H (H-2′a); 3.66 ddd, 1H, J = 13.1, 10.1 and 6.4 (H-2′b); 3.44 dq, 1H, J = 7.0 and 5.1 (H-3′); 2.00 ddd, 1H, J = 18.8, 15.3 and 5.7 (H-4′a); 1.84 ddd, 1H, J = 17.8, 15.3 and 7.0 (H-4′b); 1.17 dt, 6H, J = 7.0 and 4.0 (CH3); 1.12 d, 3H, J = 6.1 (H-5′). ^{13}C NMR (DMSO-d6): 159.63 (C-2); 153.99 (C-4); 149.13 (C-6); 143.51 (C-8); 123.10 (C-5); 76.73 (C-3′); 65.17 (C-2′); 60.75 d, 2C, J(P,C) = 6.3 ($POCH_2$); 43.13 (C-1′); 32.17 d, J(P,C) = 135.3 (C-4′), 20.57 d, J(P,C) = 8.1 (C-5′); 16.09 d, J(P,C) = 5.8 (CH_3). MS (ESI): m/z = 392–394 $[M+H]^+$.

5.8.5. Diethyl 9-[2-(1-phosphonobutan-2-yloxy)ethyl]-2-amino-6-chloropurine (26b)

Starting from the hydroxy derivative **24b**, yield 36%. ^{1}H NMR (DMSO-d6): 8.31 s, 1H (H-8); 6.90 s, 2H (NH_2); 4.18 t, 2H, $J(1',2')$ = 5.3 (H-1′); 3.92 m, 4H ($POCH_2$); 3.81 td, 1H, J = 10.1 and 5.0 (H-2′a); 3.71 td, 1H, J = 10.5 and 5.4 (H-2′b); 3.48 m, 1H (H-3′); 1.89 dddd, 2H, J = 42.6, 18.1, 15.4 and 6.3 (H-4′); 1.54 dtd, 1H, J = 12.2, 7.3 and 5.0 (H-5′a); 1.42 dt, 1H, J = 13.9 and 6.9 (H-5′b);1.18 dt, 6H, J = 7.1 and 3.1 (CH3); 0.67 t, 3H, J = 7.4 (H-6′). ^{13}C NMR (DMSO-d6): 159.61 (C-2); 154.00 (C-4); 149.10 (C-6); 143.52 (C-8); 123.12 (C-5); 75.45 (C-3′); 65.58 (C-2′); 60.76 d, 2C, J(P,C) = 6.3 ($POCH_2$); 43.22 (C-1′); 29.45 d, J(P,C) = 135.5 (C-4′), 26.79 d, J(P,C) = 8.5 (C-5′); 16.10 d, J(P,C) = 5.8 (CH_3); 8.58 (C-6′). MS (ESI): m/z = 406–408 $[M+H]^+$.

5.8.6. Diethyl 9-[2-(3-phenyl-1-phosphonopropan-2-yloxy)ethyl]-2-amino-6-chloropurine (26c)

Starting from the hydroxy derivative **24c**, yield 74%. ^{1}H NMR (DMSO-d6): 7.98 s, 1H (H-8); 7.13 m, 3H and 7.02 m, 2H (Ar); 6.88 s, 2H (NH_2); 4.11 t, 2H, $J(1',2')$ = 4.8 (H-1′); 3.90 m, 4H ($POCH_2$); 3.81 td, 1H, J = 4.8 and 10.0, 3.75 m, 1H and 3.65 td, 1H, J = 5.4 and 10.6 (H-2′ and H-3′); 2.82 dd, 1H, $J(5'b,3')$ = 4.9, J_g = 13.7 (H-5′a); 2.71 dd, 1H, $J(5'b,3')$ = 6.8, J_g = 13.7 (H-5′b); 1.89 ddt, 2H, J = 6.2, 15.5, 21.3 (H-4′); 1.17 dt, 6H, J = 7.0 and 1.3 (CH3). ^{13}C NMR (DMSO-d_6): 159.91 (C-2); 154.26 (C-4); 149.43 (C-6); 143.76 (C-8); 138.08 (Ar); 129.50, 2C (Ar); 128.21, 2C (Ar); 126.28 (Ar); 123.53 (C-5); 76.05 d, J(P,C) = 2.0 (C-3′); 66.43 (C-2′); 61.15 d, 2C, J(P,C) = 6.2 ($POCH_2$); 43.51 (C-1′); 40.60 d, J(P,C) = 10.1 (C-5′); 30.05 d, J(P,C) = 136.3 (C-4′), 16.44 d, J(P,C) = 5.8 (CH_3). MS (ESI): m/z = 468–470 $[M+H]^+$.

5.9. Synthesis of 9-[2-(2-phosphonoalkoxy)ethyl]adenines 12—general procedure

A mixture of diethyl esters **11a**–**11c** (1 mmol), acetonitrile (20 ml) and $BrSiMe_3$ (2 ml) was stirred overnight at room temper-

ature. After evaporation and codistillation with acetonitrile, the residue was treated with aqueous ammonia (2.5%) and evaporated to dryness. The residue was applied onto a column of Dowex 50 x 8 (H^+-form, 40 ml) and washed with water. Elution with 2.5% aqueous ammonia and evaporation afforded crude product as the ammonium salt. This residue was applied onto a column of Dowex 1 x 2 (acetate, 50 ml), washed with water followed by a gradient of acetic acid (0–0.5 M). The main UV-absorbing fraction was evaporated and codistilled with water to obtain product as a white solid.

5.9.1. 9-[2-(2-Phosphonopropoxy)ethyl]adenine (12a)

Starting from diester **11a**, yield 78%. ^{1}H NMR (DMSO-d_6): 7.97 s, 1H and 7.95 s, 1H (H-2, H-8); 4.25 ddd, 1H and 4.18, J(1′,2′) = 3.9 and 6.5, J_g = 14.9 (H-1′); 3.79 ddd, 1H, and 3.64J(2′,1′) = 3.9 and 6.5, J_g = 11.4 (H-2′); 3.71 dt, 1H, J(3′a,4′) = J(3′a,P) = 3.6, J_g = 10.0 (H-3′a); 3.16 ddd, 1H, J(3′b,4′) = 11.7, J(3′b,P) = 2.9, J_g = 10.0 (H-3′b); 1.56 m, 1H (H-4′); 0.68 dd, 3H, J(5′,4′) = 6.9, J(5′,P) = 15.6 (H-5′). ^{13}C NMR (DMSO-d_6): 156.91 (C-6); 148.65 (C-4); 145.68 (C-2); 140.93 (C-8); 124.01 (C-5); 71.10 d, J(P,C) = 3.6 (C-3′); 68.33 (C-2′); 43.41 (C-1′); 31.12 d, J(P,C) = 135.6 (C-4′); 11.91 d, J(P,C) = 5.1 (C-5′). MS (ESI): m/z = 300 [M−H]$^-$. Anal. Calcd for $C_{10}H_{16}N_5O_4P$: C, 39.87; H, 5.35; N, 23.25. Found: C, 39.70; H, 5.27; N, 23.07.

5.9.2. 9-[2-(2-Phosphonobutoxy)ethyl]adenine (12b)

Starting from diester **11b**, yield 66%. ^{1}H NMR (DMSO-d_6): 8.16 s, 1H and 8.12 s, 1H (H-2, H-8); 7.38 br s, 2H (NH_2); 4.30 t, 2H, J(1′,2′) = 5.3 (H-1′); 3.75 dt, 1H, and 3.70 dt, 1H, J(2′,1′) = 5.3, J_g = 10.6 (H-2′); 3.64 ddd, 1H, J(3′a,4′) = 3.9, J_g = 9.6, J(3′a,P) = 10.4 (H-3′a); 3.39 ddd, 1H, J(3′b,4′) = 6.8, J_g = 9.6, J(3′b,P) = 9.0 (H-3′b); 1.66 m, 1H (H-4′); 1.45 m, 2H (H-5′); 0.79 t, 3H, J(6′,5′) = 7.5 (H-6′). ^{13}C NMR (DMSO-d_6): 155.36 (C-6); 151.58 (C-2); 149.35 (C-4); 141.48 (C-8); 118.48 (C-5); 69.22 d, J(P,C) = 2.5 (C-3′); 68.09 (C-2′); 42.84 (C-1′); 39.46 d, J(P,C) = 134.0 (C-4′); 19.51 d, J(P,C) = 3.6 (C-5′); 12.08 d, J(P,C) = 7.8 (C-6′). MS (ESI): m/z = 316 [M−H]$^-$. Anal. Calcd for $C_{11}H_{18}N_5O_4P{\cdot}H_2O$: C, 39.64; H, 6.05; N, 21.01. Found: C, 39.59; H, 5.88; N, 20.89.

5.9.3. 9-[2-(3-Phenyl-2-phosphonopropoxy)ethyl]adenine (12c)

Starting from diester **11c**, yield 74%. ^{1}H NMR (DMSO-d_6): 8.14 s, 1H and 8.04 s, 1H (H-2, H-8); 7.27 br s, 2H (NH_2); 7.10 m, 3H and 6.93 m, 2H (Ar); 4.25 m, 2H (H-1′); 3.73 dt, 1H and 3.67 dt, 1H, J(2′,1′) = 4.6 and 5.7, J_g = 10.3 (H-2′); 3.41 m, 2H (H-3′); 2.88 td, 1H, J(5′a,4′) = 4.7, J(5′a,P) = J_g = 13.8 (H-5′a); 2.60 d br t, 1H, J(5′b,4′) = J(5′a,P) = 9.5, J_g= 13.8 (H-5′b); 1.98 m, 1H (H-4′). ^{13}C NMR (DMSO-d_6): 156.01 (C-6); 152.31 (C-2); 149.59 (C-4); 141.65 (C-8); 140.37 d, J(P,C) = 11.8 (Ar); 129.05, 2C, 128.13, 2C and 125.91 (Ar); 118.77 (C-5); 68.63 (C-3′); 68.43 (C-2′); 43.05 (C-1′); 40.21 d, J(P,C) = 134.4 (C-4′); 32.25 d, J(P,C) = 1.4 (C-5′). MS (ESI): m/z = 376 [M−H]$^-$. Anal. Calcd for $C_{16}H_{20}N_5O_4P{\cdot}H_2O$: C, 48.61; H, 5.61; N, 17.71. Found: C, 48.64; H, 5.22; N, 17.39.

5.10. Synthesis of 9-[2-(phosphonoalkoxy)ethyl]-6-chloropurines 15 and 27—general procedure

A mixture of diethyl ester **13** or **25** (0.5 mmol), acetonitrile (7 ml), $BrSiMe_3$ (0.5 ml) and 2,6-lutidine (0.4 ml) was stirred overnight at room temperature. After evaporation and codistillation with acetonitrile (5×), the residue was treated with aqueous methanol (2:1, 20 ml) for 0.5 h, evaporated and codistilled with water. The residue was purified by preparative HPLC (water–methanol) and the product was obtained after codistillation with methanol as colorless foam.

5.10.1. 9-[2-Phosphonopropoxy)ethyl]-6-chloropurine (15a)

Starting from diester **13a**, yield 67%. ^{1}H NMR (DMSO-d_6): 8.78 s, 1H and 8.68 s, 1H (H-2 and H-8); 4.47 m, 2H (H-1′); 3.83 ddd, 1H, J(2′a,1′) = 4.7 and 5.6, J_g = 10.9 (H-2′a) 3.75 ddd, 1H, J(2′b,1′) = 4.6 and 6.0, J_g = 10.9 (H-2′b); 3.64 ddd, 1H, J(3′a,4′) = 4.0, J_g = 9.6, J(3′a,P) = 7.5 (H-3′a); 3.26 td, 1H, J(3′b,4′) = J_g = 9.7, J(3′b,P) = 6.3, (H-3′b); 1.81 m, 2H (H-4′); 0.90 dd, 3H, J(5′,4′) = 7.1, J(5′,P) = 17.3 (H-5′). ^{13}C NMR (DMSO-d_6): 151.90 (C-4); 151.36 (C-2); 148.80 (C-6); 147.79 (C-8); 130.58 (C-5); 70.70 d. J(P,C) = 3.3 (C-3′); 67.51 (C-2′); 43.52 (C-1′); 32.69 d, J(P,C) = 135.4 (C-4′); 11.51 d, J(P,C) = 4.9 (C-5′). MS (ESI): m/z = 319–321 [M−H]$^-$. Anal. Calcd for $C_{10}H_{14}ClN_4O_4P{\cdot}3/5MeOH$: C, 37.46; H, 4.87; N, 16.48. Found: C, 37.49; H, 4.64; N, 16.18.

5.10.2. 9-[2-(3-Phenyl-2-phosphonopropoxy)ethyl]-6-chloropurine (15c)

Starting from diester **13c**, yield 48%. ^{1}H NMR (DMSO-d_6): 8.78 s, 1H and 8.62 s, 1H (H-2 and H-8); 7.09 m, 3H and 6.90 m, 2H (Ar); 4.41 br t, 2H, J(1′,2′) = 4.7 and 5.6 (H-1′); 3.68 d br t, 1H, and 3.63 d br t, 1H, J(2′,1′) = 4.7 and 5.6, J_g = 10.7 (H-2′); 3.42 m, 2H (H-3′); 2.86 td, 1H, J(5′a,4′) = 4.6, J_g = J(5′a,P) = 13.6 (H-5′a); 2.56 d br t, 1H, J(5′b,4′) = J(5′b,P) = 9.4, J_g = 13.6 (H-5′b); 1.97 m, 1H (H-4′). ^{13}C NMR (DMSO-d_6): 151.83 (C-4); 151.32 (C-2); 148.80 (C-6); 147.91 (C-8); 139.98 d, J(P,C) = 12.2 (Ar); 130.61 (C-5); 128.61, 2C (Ar); 127.81, 2C (Ar); 125.64 (Ar); 68.31 (C-3′); 67.68 (C-2′); 43.53 (C-1′); 40.30 d, J(P,C) = 134.0 (C-4′); 31.91 d, J(P,C) = 1.0 (C-5′). MS (ESI): m/z = 395–397 [M−H]$^-$. Anal. Calcd for $C_{16}H_{18}ClN_4O_4P{\cdot}1/3H_2O$: C, 47.71; H, 4.67; N, 13.91. Found: C, 47.68; H, 4.61; N, 13.70.

5.10.3. 9-[2-(1-Phosphonopropan-2-yloxy)ethyl]-6-chloropurine (27a)

Starting from diester **25a**, yield 70%. ^{1}H NMR (DMSO-d_6): 8.78 s, 1H and 8.70 s, 1H (H-2 and H-8); 4.43 t, 2H, J(1′,2′) = 5.3 (H-1′); 3.79 t, 2H, J(2′,1′) = 5.3 (H-2′); 3.65 m, 1H (H-3′); 1.84 ddd, 1H, J(4′a,3′) = 4.5, J_g = 14.7, J(4′a,P) = 19.6 (H-4′a); 1.58 ddd, 1H, J(4′b,3′) = 8.4, J_g = 14.7, J(4′b,P) = 17.6 (H-4′b); 1.11 d, 3H, J(5′,3′) = 6.1 (H-5′). ^{13}C NMR (DMSO-d_6): 151.93 (C-4); 151.39 (C-2); 148.84 (C-6); 147.91 (C-8); 130.65 (C-5); 71.43 (C-3′); 64.99 (C-2′); 43.98 (C-1′); 35.25 d, J(P,C) = 132.6 (C-4′); 20.78 d, J(P,C) = 5.4 (C-5′). MS (ESI): m/z = 319–321 [M−H]$^-$. Anal. Calcd for $C_{10}H_{14}ClN_4O_4P{\cdot}1/3H_2O$: C, 36.77; H, 4.53; N, 17.15. Found: C, 37.01; H, 4.64; N, 16.79.

5.10.4. 9-[2-(1-Phosphonobutan-2-yloxy)ethyl]-6-chloropurine (27b)

Starting from diester **25b**, yield 63%. ^{1}H NMR (DMSO-d_6): 8.89 s, 1H and 8.80 s, 1H (H-2 and H-8); 4.55 t, 2H, J(1′,2′) = 5.3 (H-1′); 3.89 dt, 1H, J(2′a,1′) = 4.8, J_g = 10.6 (H-2′a); 3.74 dt, 1H, J(2′b,1′) = 5.6, J_g = 10.6 (H-2′b); 3.46 m, 1H (H-3′); 1.78 ddd, 1H, J(4′a,3′) = 4.9, J_g = 14.9, J(4′a,P) = 19.6 (H-4′a); 1.59 m, 2H (H-4′b and H-5′a); 1.35 sept, 1H, J(5′b,3′) = J(5′b,6′) = 7.2, J_g = 14.2 (H-5′b); 0.69 t, 3H, J(6′,5′) = 7.3 (H-6′). ^{13}C NMR (DMSO-d_6): 152.16 (C-4); 151.56 (C-2); 149.01 (C-6); 148.12 (C-8); 130.88 (C-5); 76.40 (C-3′); 65.59 (C-2′); 44.30 (C-1′); 32.59 d, J(P,C) = 132.9 (C-4′); 27.17 d, J(P,C) = 5.9 (C-5′); 8.91 (C-6′). MS (ESI): m/z = 333–335 [M−H]$^-$. Anal. Calcd for $C_{11}H_{16}ClN_4O_4P{\cdot}1/5H_2O$: C, 39.05; H, 4.89; N, 16.56. Found: C, 39.16; H, 5.06; N, 16.43.

5.10.5. 9-[2-(3-Phenyl-1-phosphonopropan-2-yloxy)ethyl]-6-chloropurine (27c)

Starting from diester **25c**, yield 40%. ^{1}H NMR (DMSO-d_6): 8.73 s, 1H and 8.52 s, 1H (H-2 and H-8); 7.02 m, 3H and 6.91 m, 2H (Ar); 4.50 ddd, 1H, J(1′,2′) = 4.0 and 7.3, J_g = 14.2 (H-1′a); 4.45 dt, 1H, J(1′,2′) = 4.0, J_g = 14.2 (H-1′b); 3.99 br dt, 1H, J(2′,1′) = 4.4, J_g = 10.5 (H-2′a); 3.75 ddd, 1H, J(2′,1′) = 7.3 and 4.0, J_g = 10.5 (H-2′b);

3.85 m, 1H (H-3′); 3.00 dd, 1H, J(5′a,3′) = 4.1, J_g = 13.9 (H-5′a); 2.71 dd, 1H, J(5′b,3′) = 7.5, J_g = 13.9 (H-5′b); 1.93 ddd, 1H, J(4′a,3′) = 5.3, J_g = 14.9, J(4′a,P) = 19.1 (H-4′a); 1.77 ddd, 1H, J(4′b,3′) = 7.4, J_g = 14.9, J(4′b,P) = 17.5 (H-4′b). ^{13}C NMR (DMSO-d_6): 151.75 (C-4); 151.14 (C-2); 148.71 (C-6); 147.66 (C-8); 138.18 (Ar); 130.67 (C-5); 128.93, 2C (Ar); 127.54, 2C (Ar); 125.58 (Ar); 76.25 (C-3′); 65.70 (C-2′); 43.99 (C-1′); 40.46 d, J(P,C) = 7.3 (C-5′); 32.53 d, J(P,C) = 133.0 (C-4′). MS (ESI): m/z = 395–397 [M−H]$^-$. Anal. Calcd for $C_{16}H_{18}ClN_4O_4P\cdot 1/3H_2O$: C, 47.71; H, 4.67; N, 13.91. Found: C, 47.68; H, 4.61; N, 13.70.

5.11. Synthesis of 9-[2-(phosphonoalkoxy)ethyl]-2-amino-6-chloropurines 16 and 28—general procedure

Prepared as described above for the 6-chloropurine derivatives starting from diethyl esters **14** or **26** (0.5 mmol). After preparative HPLC and codistillation with methanol products were obtained as colorless foams.

5.11.1. 9-[2-Phosphonopropoxy)ethyl]-2-amino-6-chloropurine (16a)

Starting from diester **14a**, yield 65%. ^{1}H NMR (DMSO-d_6): 8.09 s, 1H (H-8); 6.90 br s, 2H (NH_2); 4.20 t, 2H (H-1′); 3.73 m, 1H (H-3′a); 3.67 m, 2H (H-2′); 3.25 m, 1H (H-3′b); 1.83 m, 1H (H-4′);0.96 d, 3H, J(5′,3′) = 6.0 (H-5′). ^{13}C NMR (DMSO-d_6): 159.81 (C-2); 153.65 (C-4); 149.22 (C-6); 143.92 (C-8); 123.16 (C-5);70.51 d, J(P,C) = 3.5 (C-3′); 67.81 (C-2′); 42.76 (C-1′); 32.53 d, J(P,C) = 135.0 (C-4′); 11.55 d, J(P,C) = 4.9 (C-5′). MS (ESI): m/z = 334–336 [M−H]$^-$. Anal. Calcd for $C_{10}H_{15}ClN_5O_4P$: C, 35.78; H, 4.50; N, 20.86. Found: C, 35.65; H, 4.62; N, 20.59.

5.11.2. 9-[2-(3-Phenyl-2-phosphonopropoxy)ethyl]-2-amino-6-chloropurine (16c)

Starting from diester **14c**, yield 36%. ^{1}H NMR (DMSO-d_6): 8.03 s, 1H (H-8); 7.24 m, 3H and 7.05 m, 2H (Ar); 7.02 br s, 2H (NH_2); 4.15 br t, 2H, J(1′,2′) = 4.9 and 5.5 (H-1′); 3.71 d br t, 1H, and 3.67 d br t, 1H J(2′,1′) = 4.9 and 5.5, J_g = 10.2 (H-2′); 3.52 m, 2H (H-3′); 2.98 td, 1H, J(5′a,4′) = 4.7, J_g = J(5′a,P) = 13.8 (H-5′a); 2.70 d br t, 1H, J(5′b,4′) = J(5′b,P) = 9.5, J_g = 13.7 (H-5′b); 2.10 m, 1H (H-4′). ^{13}C NMR (DMSO-d_6): 159.69 (C-2); 153.95 (C-4); 149.16 (C-6); 143.67 (C-8); 140.01 d, J(P,C) = 11.9 (Ar); 128.83, 2C (Ar); 127.84, 2C (Ar); 125.65 (Ar); 123.13 (C-5); 68.28 (C-3′); 67.68 (C-2′); 42.77 (C-1′); 40.18 d, J(P,C) = 134.9 (C-4′); 31.93 d, J(P,C) = 2.0 (C-5′). MS (ESI): m/z = 410–412 [M−H]$^-$. Anal. Calcd for $C_{16}H_{19}ClN_5O_4P\cdot 3/4H_2O$: C, 45.19; H, 4.86; N, 16.47. Found: C, 45.49; H, 4.89; N, 16.16.

5.11.3. 9-[2-(1-Phosphonopropan-2-yloxy)ethyl]-2-amino-6-chloropurine (28a)

Starting from diester **26a**, yield 55%. ^{1}H NMR (DMSO-d_6): 8.10 s, 1H (H-8); 6.90 br s, 2H (NH_2); 4.16 t, 2H, J(1′,2′) = 5.4 (H-1′); 3.70 t, 2H, J(2′,1′) = 5.4 (H-2′); 3.65 m, 1H (H-3′); 1.87 ddd, 1H, J(4′a,3′) = 4.1, J_g = 14.7, J(4′a,P) = 19.8 (H-4′a); 1.59 ddd, 1H, J(4′b,3′) = 8.7, J_g = 14.7, J(4′b,P) = 17.4 (H-4′b); 1.13 d, 3H, J(5′,3′) = 6.1 (H-5′). ^{13}C NMR (DMSO-d_6): 159.41 (C-2); 153.76 (C-4); 148.90 (C-6); 143.32 (C-8); 122.85 (C-5); 71.13 (C-3′); 64.77 (C-2′); 42.86 (C-1′); 35.33 d, J(P,C) = 132.7 (C-4′); 20.56 d, J(P,C) = 5.0 (C-5′). MS (ESI): m/z = 334–336 [M−H]$^-$. Anal. Calcd for $C_{10}H_{15}ClN_5O_4P\cdot 1/5H_2O$: C, 35.40; H, 4.57; N, 20.64. Found: C, 35.33; H, 4.46; N, 20.54.

5.11.4. 9-[2-(1-Phosphonobutan-2-yloxy)ethyl]-2-amino-6-chloropurine (28b)

Starting from diester **26b**, yield 37%. ^{1}H NMR (DMSO-d_6): 8.10 s, 1H (H-8); 6.90 br s, 2H (NH_2); 4.18 t, 2H, J(1′,2′) = 5.5 (H-1′); 3.80 dt, 1H, J(2′a,1′) = 5.0, J_g = 10.4 (H-2′a); 3.65 dt, 1H, J(2′b,1′) = 6.1, J_g = 10.4 (H-2′b); 3.45 m, 1H (H-3′); 1.80 ddd, 1H, J(4′a,3′) = 4.7, J_g = 14.9, J(4′a,P) = 19.7 (H-4′a); 1.65 ddd, 1H, J(4′b,3′) = 7.9, J_g = 14.9, J(4′b,P) = 17.6 (H-4′b); 1.63 m, 1H, J(5′a,3′) = 4.0, J(5′a,6′) = 7.3, J_g = 14.2 (H-5′a); 1.38 sept, 1H, J(5′b,3′) = J(5′b,6′) = 7.3, J_g = 14.2 (H-5′b); 0.65 t, 3H, J(5′,3′) = 7.3 (H-6′). ^{13}C NMR (DMSO-d_6): 159.88 (C-2); 154.28 (C-4); 149.36 (C-6); 143.84 (C-8); 123.38 (C-5); 76.38 (C-3′); 65.68 (C-2′); 43.46 (C-1′); 32.16 d, J(P,C) = 132.6 (C-4′); 27.21 d, J(P,C) = 5.6 (C-5′); 9.00 (C-6′). MS (ESI): m/z = 348–350 [M−H]$^-$. Anal. Calcd for $C_{11}H_{17}ClN_5O_4P\cdot 2/5H_2O$: C, 37.02; H, 5.03; N, 19.62. Found: C, 37.26; H, 5.15; N, 19.30.

5.11.5. 9-[2-(3-Phenyl-1-phosphonopropan-2-yloxy)ethyl]-2-amino-6-chloropurine (28c)

Starting from diester **26c**, yield 60%. ^{1}H NMR (DMSO-d_6): 7.95 s, 1H (H-8); 7.10 m, 3H and 7.00 m, 2H (Ar); 6.87 br s, 2H (NH_2); 4.10 m, 2H (H-1′); 3.70 m, 1H (H-2′a); 3.58 ddd, 1H, J(2′,1′) = 7.5 and 3.4, J_g = 10.6 (H-2′b); 3.76 m, 1H (H-3′); 2.91 dd, 1H, J(5′a,3′) = 4.1, J_g = 14.0 (H-5′a); 2.65 dd, 1H, J(5′b,3′) = 7.2, J_g = 14.0 (H-5′b); 1.81 ddd, 1H, J(4′a,3′) = 5.1, J_g = 14.9, J(4′a,P) = 19.2 (H-4′a); 1.67 ddd, 1H, J(4′b,3′) = 7.4, J_g = 14.9, J(4′b,P) = 17.4 (H-4′b). ^{13}C NMR (DMSO-d_6): 159.55 (C-2); 153.92 (C-4); 149.06 (C-6); 143.46 (C-8); 138.22 (Ar); 129.18, 2C (Ar); 127.70, 2C (Ar); 125.72 (Ar); 123.18 (C-5); 76.22 (C-3′); 65.84 (C-2′); 43.15 (C-1′); 40.38 d, J(P,C) = 7.3 (C-5′); 32.49 d, J(P,C) = 133.0 (C-4′). MS (ESI): m/z = 410–412 [M−H]$^-$. Anal. Calcd for $C_{16}H_{19}ClN_5O_4P\cdot 5/6H_2O$: C, 45.03; H, 4.88; N, 16.41. Found: C, 45.17; H, 4.71; N, 16.00.

5.12. Synthesis of 9-[2-(phosphonoalkoxy)ethyl]-6-bromopurines 17 and 29—general procedure

A mixture of diethyl ester **13** or **25** (2 mmol), acetonitrile (20 ml) and $BrSiMe_3$ (2 ml) was stirred overnight at room temperature. After evaporation and codistillation with acetonitrile, the residue was treated with aqueous methanol (2:1, 60 ml) for 1 h, evaporated and codistilled with water. The residue was purified by preparative HPLC (water–methanol) and solid yellowish product was obtained after lyophilization.

5.12.1. 9-[2-(2-Phosphonopropoxy)ethyl]-6-bromopurine (17a)

Starting from diester **13a**, yield 80%. ^{1}H NMR (DMSO-d_6): 8.73 s, 1H and 8.68 s, 1H (H-2, H-8); 4.57 m, 2H (H-1′); 3.87 ddd, 1H, and 3.74 ddd, 1H J(2′,1′) = 4.6 and 6.0, J_g = 10.6 (H-2′); 3.635 ddd, 1H, J(3′a,4′) = 4.0, J(3′a,P) = 7.6, J_g = 9.6 (H-3′a); 3.26 td, 1H, J(3′b,P) = 6.3, J(3′b,4′) = J_g = 9.7 (H-3′b); 1.81 m, 1H, J(4′,P) = 20.6, (H-4′); 0.90 dd, 3H, J(5′,4′) = 7.0, J(5′,P) = 17.3 (H-5′). ^{13}C NMR (DMSO-d_6): 151.64 (C-2); 150.91 (C-4); 147.85 (C-8); 141.65 (C-6); 133.46 (C-5); 70.97 d, J(P,C) = 3.3 (C-3′); 67.77 (C-2′); 43.79 (C-1′); 32.94 d, J(P,C) = 135.2 (C-4′); 11.76 d, J(P,C) = 4.9 (C-5′). MS (ESI): m/z = 363–365 [M−H]$^-$. Anal. Calcd for $C_{10}H_{14}BrN_4O_4P$: C, 32.90; H, 3.86; N, 15.34. Found: C, 32.93; H, 3.76; N, 15.43.

5.12.2. 9-[2-(2-Phosphonobutoxy)ethyl]-6-bromopurine (17b)

Starting from diester **13b**, yield 82%. ^{1}H NMR (DMSO-d_6): 8.72 s, 1H and 8.68 s, 1H (H-2, H-8); 4.46 br t, 2H, J(1′,2′) = 5.4 (H-1′); 3.91 ddd, 1H, and 3.86 ddd, 1H, J(2′,1′) = 4.7 and 5.8, J_g = 10.9 (H-2′); 3.74 ddd, 1H, J(3′a,4′) = 4.0, J_g = 9.8, J(3′a,P) = 11.1 (H-3′a); 3.50 ddd, 1H, J(3′b,4′) = 8.7, J_g = 9.8, J(3′b,P) = 7.2 (H-3′b); 1.62 m, 1H (H-4′); 1.45 m, 1H and 1.32 m, 1H (H-5′); 0.72 t, 3H, J(6′,5′) = 7.5 (H-6′). ^{13}C NMR (DMSO-d_6): 151.33 (C-2); 150.65 (C-4); 147.59 (C-8); 141.36 (C-6); 133.21 (C-5); 69.06 d, J(P,C) = 2.7 (C-3′); 67.64 (C-2′); 43.55 (C-1′); 39.91 d, J(P,C) = 133.7 (C-4′); 19.35 d, J(P,C) = 3.4 (C-5′); 11.86 d, J(P,C) = 8.0 (C-6′). MS (ESI): m/z = 377–379 [M−H]$^-$. Anal. Calcd for $C_{11}H_{16}BrN_4O_4P$: C, 34.85; H, 4.25; N, 14.78. Found: C, 35.16; H, 4.34; N, 14.56.

5.12.3. 9-[2-(3-Phenyl-2-phosphonopropoxy)ethyl]-6-bromopurine (17c)

Starting from diester **13c**, yield 87%. ^{1}H NMR (DMSO-d_6): 8.72 s, 1H and 8.63 s, 1H (H-2, H-8); 7.10 m, 3H and 6.90 m, 2H (Ar); 4.40 m, 2H (H-1′); 3.68 ddd, 1H, and 3.60 ddd, 1H J(2′,1′) = 4.5 and 5.9, J_g = 10.4 (H-2′); 3.40 m, 2H (H-3′); 1.98 m, 1H (H-4′); 2.86 td, 1H, J(5′a,4′) = 4.6, J_g = J(5′a,P) = 13.7 (H-5′a); 2.56 dd, 1H, J(5′b,4′) = J(5′b,P) = 9.7, J_g = 13.7 (H-5′b). ^{13}C NMR (DMSO-d_6): 151.90 (C-2); 151.13 (C-4); 148.27 (C-8); 141.96 (C-6); 140.52 d, J(P,C) = 12.1 (Ar); 129.17, 2C (Ar); 128.38, 2C (Ar); 126.21 (Ar); 133.79 (C-5); 68.83 d, J(P,C) = 1.5 (C-3′); 68,23 (C-2′); 44.09 (C-1′); 40.61 d, J(P,C) = 134.5 (C-4′); 32.46 d, J(P,C) = 4.9 (C-5′). MS (ESI): m/z = 439–441 [M−H]$^-$. Anal. Calcd for $C_{16}H_{18}BrN_4O_4P$: C, 43.55; H, 4.11; N, 12.70. Found: C, 43.36; H, 4.36; N, 12.30.

5.12.4. 9-[2-(1-Phosphonopropan-2-yloxy)ethyl]-6-bromopurine (29a)

Starting from diester **25a**, yield 70%. ^{1}H NMR (DMSO-d_6): 8.72 s, 1H and 8.69 s, 1H (H-2 and H-8); 4.42 t, 2H, J(1′,2′) = 5.2 (H-1′); 3.79 t, 2H, J(2′,1′) = 5.2 (H-2′); 3.60 m, 1H (H-3′); 1.84 ddd, 1H, J(4′a,3′) = 4.4, J_g = 14.7, J(4′a,P) = 19.5 (H-4′a); 1.57 ddd, 1H, J(4′b,3′) = 8.5, J_g = 14.7, J(4′b,P) = 17.5 (H-4′b); 1.11 d, 3H, J(5′,3′) = 6.1 (H-5′). ^{13}C NMR (DMSO-d_6): 151.40 (C-2); 150.67 (C-4); 147.70 (C-8); 141.42 (C-6); 133.25 (C-5); 71.44 (C-3′); 64.97 (C-2′); 43.98 (C-1′); 35.27 d, J(P,C) = 132.9 (C-4′); 20.80 d, J(P,C) = 5.3 (C-5′). MS (ESI): m/z = 363–365 [M−H]$^-$. Anal. Calcd for $C_{10}H_{14}BrN_4O_4P\cdot 1/3MeOH$: C, 33.03; H, 4.11; N, 14.91. Found: C, 33.07; H, 4.12; N, 14.81.

5.12.5. 9-[2-(1-Phosphonobutan-2-yloxy)ethyl]-6-bromopurine (29b)

Starting from diester **25b**, yield 71%. ^{1}H NMR (DMSO-d_6): 8.72 s, 1H and 8.70 s, 1H (H-2 and H-8); 4.43 t, 2H, J(1′,2′) = 5.2 (H-1′); 3.89 dt, 2H, J(2′a,1′) = 4.9, J_g = 10.6 (H-2′a); 3.74 dt, 2H, J(2′b,1′) = 5.5, J_g = 10.6 (H-2′b); 3.46 m, 1H (H-3′); 1.78 ddd, 1H, J(4′a,3′) = 4.9, J_g = 14.9, J(4′a,P) = 19.6 (H-4′a); 1.60 m, 2H (H-4′b and H-5′a); 1.35 sept, 1H, J(5′b,3′) = J(5′b,6′) = 7.1, J_g = 14.2 (H-5′b); 0.58 t, 3H, J(6′,5′) = 7.3 (H-6′). ^{13}C NMR (DMSO-d_6): 151.58 (C-2); 150.90 (C-4); 147.92 (C-8); 141.60 (C-6); 133.49 (C-5); 76.40 (C-3′); 65.59 (C-2′); 44.31 (C-1′); 32.59 d, J(P,C) = 132.7 (C-4′); 27.17 d, J(P,C) = 6.1 (C-5′); 8.92 (C-6′). MS (ESI): m/z = 377–379 [M−H]$^-$. Anal. Calcd for $C_{11}H_{16}BrN_4O_4P\cdot 1/4H_2O$: C, 34.44; H, 4.33; N, 14.60. Found: C, 34.53; H, 4.52; N, 14.30.

5.12.6. 9-[2-(3-Phenyl-1-phosphonopropan-2-yloxy)ethyl]-6-bromopurine (29c)

Starting from diester **25c**, yield 61%. ^{1}H NMR (DMSO-d_6): 8.67 s, 1H and 8.52 s, 1H (H-2 and H-8); 7.02 m, 3H and 6.90 m, 2H (Ar); 4.48 ddd, 1H, J(1′,2′) = 4.0 and 7.3, J_g = 14.6 (H-1′a); 4.43 dt, 1H, J(1′,2′) = 4.5, J_g = 14.6 (H-1′b); 3.99 br dt, 1H, J(2′,1′) = 4.0 and 4.8, J_g = 10.3 (H-2′a); 3.75 ddd, 1H, J(2′,1′) = 7.3 and 4.2, J_g = 10.3 (H-2′b); 3.85 m, 1H (H-3′); 2.99 dd, 1H, J(5′a,3′) = 4.1, J_g = 13.9 (H-5′a); 2.71 dd, 1H, J(5′b,3′) = 7.4, J_g = 13.9 (H-5′b); 1.93 ddd, 1H, J(4′a,3′) = 5.2, J_g = 14.9, J(4′a,P) = 19.1 (H-4′a); 1.77 ddd, 1H, J(4′b,3′) = 7.4, J_g = 14.9, J(4′b,P) = 17.5 (H-4′b). ^{13}C NMR (DMSO-d_6): 151.18 (C-2); 150.53 (C-4); 147.50 (C-8); 141.37 (C-6); 138.21 (Ar); 133.31 (C-5); 128.96, 2C (Ar); 127.58, 2C (Ar); 125.62 (Ar); 76.28 (C-3′); 65.73 (C-2′); 44.03 (C-1′); 40.49 d, J(P,C) = 6.6 (C-5′); 32.57 d, J(P,C) = 132.5 (C-4′). MS (ESI): m/z = 439–441 [M−H]$^-$. Anal. Calcd for $C_{16}H_{18}BrN_4O_4P\cdot 1/4MeOH$: C, 43.45; H, 4.26; N, 12.47. Found: C, 43.53; H, 4.20; N, 12.24.

5.13. Synthesis of 9-[2-(phosphonoalkoxy)ethyl]-2-amino-6-bromopurines 18 and 30—general procedure

Prepared as described above for the 6-bromopurine derivatives starting from diethyl esters **14** or **26** (3 mmol). After preparative HPLC and codistillation with methanol products were obtained as colorless foams.

5.13.1. 9-[2-(2-Phosphonopropoxy)ethyl]-2-amino-6-bromopurine (18a)

Starting from diester **14a**, yield 89%. ^{1}H NMR (DMSO-d_6): 8.09 s, 1H (H-8); 6.92 br s, 2H (NH_2); 4.19 m, 2H (H-1′); 3.75 dt, 1H, and 3.67, 1H, J(2′,1′) = 5.0, J_g = 10.7 (H-2′); 3.66 ddd, 1H, J(3′a,4′) = 4.1, J(3′a,P) = 7.3, J_g = 9.6 (H-3′a); 3.25 td, 1H, J(3′b,P) = 5.9, J(3′b,4′) = J_g = 9.8 (H-3′b); 1.83 m, 1H, J(4′,P) = 20.5, (H-4′); 0.95 dd, 3H, J(5′,4′) = 7.1, J(5′,P) = 17.3 (H-5′). ^{13}C NMR (DMSO-d_6): 159.50 (C-2); 152.78 (C-4); 143.29 (C-8); 141.78 (C-6); 125.58 (C-5); 70.73 d, J(P,C) = 3.6 (C-3′); 67.57 (C-2′); 42.70 (C-1′); 32.78 d, J(P,C) = 135.2 (C-4′); 11.60 d, J(P,C) = 5.0 (C-5′). MS (ESI): m/z = 378–380 [M−H]$^-$. Anal. Calcd for $C_{10}H_{15}BrN_5O_4P\cdot 1/2MeOH$: C, 31.83; H, 4.33; N, 17.68. Found: C, 31.58; H, 4.11; N, 17.68.

5.13.2. 9-[2-(2-Phosphonobutoxy)ethyl]-2-amino-6-bromopurine (18b)

Starting from diester **14b**, yield 79%. ^{1}H NMR (DMSO-d_6): 8.09 s, 1H (H-8); 6.91 br s, 2H (NH_2); 4.19 t, 2H, J(1′,2′) = 5.3 (H-1′); 3.73 dt, 1H, and 3.66 dt, 1H, J(2′,1′) = 5.3, J_g = 10.6 (H-2′); 3.65 ddd, 1H, J(3′a,4′) = 3.9, J_g = 9.7, J(3′a,P) = 10.6 (H-3′a); 3.38 ddd, 1H, J(3′b,4′) = 8.8, J_g = 9.7, J(3′b,P) = 6.8 (H-3′b); 1.65 m, 1H (H-4′); 1.46 m, 2H (H-5′); 0.80 t, 3H, J(6′,5′) = 7.5 (H-6′). ^{13}C NMR (DMSO-d_6): 160.05 (C-2); 153.33 (C-4); 143.87 (C-8); 142.32 (C-6); 126.16 (C-5); 69.71 d, J(P,C) = 2.5 (C-3′); 68.23 (C-2′); 43.30 (C-1′); 39.99 d, J(P,C) = 134.5 (C-4′); 19.97 d, J(P,C) = 3.4 (C-5′); 12.56 d, J(P,C) = 7.6 (C-6′). MS (ESI): m/z = 392–394 [M−H]$^-$. Anal. Calcd for $C_{11}H_{17}BrN_5O_4P$: C, 33.52; H, 4.35; N, 17.77. Found: C, 33.48; H, 4.34; N, 17.41.

5.13.3. 9-[2-(3-Phenyl-2-phosphonopropoxy)ethyl]-2-amino-6-bromopurine (18c)

Starting from diester **14c**, yield 60%. ^{1}H NMR (DMSO-d_6): 8.14 s, 1H (H-8); 7.22 m, 3H and 7.04 m, 2H (Ar); 7.03 br s, 2H (NH_2); 4.25 br t, 2H, J(1′,2′) = 4.8 and 5.9 (H-1′); 3.69 d br t, 1H, and 3.66 d br t, 1H J(2′,1′) = 4.8 and 5.9, J_g = 10.4 (H-2′); 3.40 m, 2H (H-3′); 2.88 td, 1H, J(5′a,4′) = 4.6, J_g = J(5′a,P) = 13.8 (H-5′a); 2.60 ddd, 1H, J(5′b,4′) = 9.4, J(5′b,P) = 9.8, J_g = 13.8 (H-5′b); 1.98 m, 1H (H-4′). ^{13}C NMR (DMSO-d_6): 159.78 (C-2); 152.97 (C-4); 143.75 (C-8); 142.08 (C-6); 140.34 d, J(P,C) = 11.8 (Ar); 129.00, 2C (Ar); 128.11, 2C (Ar); 126.80 (C-5); 125.90 (Ar); 68.59 br s (C-3′); 67.94 (C-2′); 43.05 (C-1′); 41.10 d, J(P,C) = 134.4 (C-4′); 32.22 br s (C-5′). MS (ESI): m/z = 454–456 [M−H]$^-$. Anal. Calcd for $C_{16}H_{19}BrN_5O_4P\cdot 1/3H_2O$: C, 41.57; H, 4.29; N, 15.15. Found: C, 41.87; H, 4.22; N, 14.94.

5.13.4. 9-[2-(1-Phosphonopropan-2-yloxy)ethyl]-2-amino-6-bromopurine (30a)

Starting from diester **26a**, yield 66%. ^{1}H NMR (DMSO-d_6): 8.10 s, 1H (H-8); 6.91 br s, 2H (NH_2); 4.15 t, 2H, J(1′,2′) = 5.4 (H-1′); 3.70 t, 2H, J(2′,1′) = 5.4 (H-2′); 3.65 m, 1H (H-3′); 1.87 ddd, 1H, J(4′a,3′) = 4.1, J_g = 14.7, J(4′a,P) = 19.8 (H-4′a); 1.59 ddd, 1H, J(4′b,3′) = 8.7, J_g = 14.7, J(4′b,P) = 17.5 (H-4′b); 1.13 d, 3H, J(5′,3′) = 6.1 (H-5′). ^{13}C NMR (DMSO-d_6): 159.46 (C-2); 153.01 (C-4); 143.33 (C-8); 141.75 (C-6); 125.54 (C-5); 71.31 (C-3′); 64.94 (C-2′); 43.05 (C-1′); 35.33 d, J(P,C) = 132.1 (C-4′); 20.74 d, J(P,C) = 5.1 (C-5′). MS (ESI): m/z = 378–380 [M−H]$^-$. Anal. Calcd for $C_{10}H_{15}BrN_5O_4P\cdot 1/3MeOH$: C, 31.76; H, 4.21; N, 17.92. Found: C, 31.73; H, 4.16; N, 17.84.

5.13.5. 9-[2-(1-Phosphonobutan-2-yloxy)ethyl]-2-amino-6-bromopurine (30b)

Starting from diester **26b**, yield 60%. ^{1}H NMR (DMSO-d_6): 8.10 s, 1H (H-8); 6.91 br s, 2H (NH_2); 4.17 t, 2H, J(1′,2′) = 5.4 (H-1′); 3.80 dt, 1H, J(2′a,1′) = 4.9, J_g = 10.5 (H-2′a); 3.65 dt, 1H, J(2′b,1′) = 6.0, J_g = 10.5 (H-2′b); 3.47 m, 1H (H-3′); 1.80 ddd, 1H, J(4′a,3′) = 4.6, J_g = 14.9, J(4′a,P) = 19.8 (H-4′a); 1.62 ddd, 1H, J(4′b,3′) = 7.9, J_g = 14.9, J(4′b,P) = 17.7 (H-4′b); 1.62 m, 1H (H-5′a); 1.38 sept, 1H, J(5′b,3′) = J(5′b,6′) = 7.1, J_g = 14.2 (H-5′b); 0.65 t, 3H, J(6′,5′) = 7.3 (H-6′). ^{13}C NMR (DMSO-d_6): 159.74 (C-2); 153.03 (C-4); 143.64 (C-8); 142.01 (C-6); 125.88 (C-5); 76.39 (C-3′); 65.67 (C-2′); 43.47 (C-1′); 32.17 d, J(P,C) = 132.6 (C-4′); 27.18 d, J(P,C) = 5.7 (C-5′); 9.00 (C-6′). MS (ESI): m/z = 392–394 $[M-H]^-$. Anal. Calcd for $C_{11}H_{17}BrN_5O_4P\cdot1/3$MeOH: C, 33.89; H, 4.70; N, 17.19. Found: C, 33.96; H, 4.71; N, 17.46.

5.13.6. 9-[2-(3-Phenyl-1-phosphonopropan-2-yloxy)ethyl]-2-amino-6-bromopurine (30c)

Starting from diester **26c**, yield 53%. ^{1}H NMR (DMSO-d_6): 7.95 s, 1H (H-8); 7.09 m, 3H and 6.97 m, 2H (Ar); 6.88 br s, 2H (NH_2); 4.14 ddd, 1H, J(1′,2′) = 4.0 and 7.4, J_g = 14.3 (H-1′a); 4.06 br dt, 1H, J(1′,2′) = 4.4, J_g = 14.3 (H-1′b); 3.78 br dt, 1H, J(2′,1′) = 4.8, J_g = 10.5 (H-2′a); 3.57 ddd, 1H, J(2′,1′) = 7.4 and 4.0, J_g = 10.5 (H-2′b); 3.75 m, 1H (H-3′); 2.91 dd, 1H, J(5′a,3′) = 4.1, J_g = 14.0 (H-5′a); 2.64 dd, 1H, J(5′b,3′) = 7.2, J_g = 14.0 (H-5′b); 1.81 ddd, 1H, J(4′a,3′) = 5.2, J_g = 14.9, J(4′a,P) = 19.2 (H-4′a); 1.66 ddd, 1H, J(4′b,3′) = 7.4, J_g = 14.9, J(4′b,P) = 17.4 (H-4′b). ^{13}C NMR (DMSO-d_6): 159.74 (C-2); 153.00 (C-4); 143.60 (C-8); 142.08 (C-6); 138.55 (Ar); 129.51, 2C (Ar); 128.05, 2C (Ar); 126.05 (Ar); 126.02 (C-5); 76.56 (C-3′); 66.17 (C-2′); 43.51 (C-1′); 40.71 d, J(P,C) = 7.3 (C-5′); 32.82 d, J(P,C) = 133.0 (C-4′). MS (ESI): m/z = 454–456 $[M-H]^-$. Anal. Calcd for $C_{16}H_{19}BrN_5O_4P\cdot1/2$MeOH: C, 41.96; H, 4.48; N, 14.83. Found: C, 42.05; H, 4.28; N, 14.58.

5.14. Synthesis of 9-[2-(phosphonoalkoxy)ethyl]hypoxanthines 19 and 31—general procedure

The 6-bromo derivative **17** or **29** (1 mmol) was dissolved in trifluoroacetic acid (75%, 16 ml) and stirred overnight. The solvent was evaporated and the residue codistilled with water (3x).After preparative HPLC and codistillation with methanol pure products were obtained.

5.14.1. 9-[2-(2-Phosphonopropoxy)ethyl]hypoxanthine (19a)

Starting from bromo derivative **17a**, colorless foam, yield 67%. ^{1}H NMR (DMSO-d_6): 12.8 br s, 1H (OH); 8.05 s, 1H and 8.03 s, 1H (H-2, H-8); 4.29 m, 2H (H-1′); 3.77 ddd, 1H, J(2′a,1′) = 4.6 and 6.0, J_g = 10.8 (H-2′a); 3.67 ddd, 1H, J(2′b,1′) = 4.8 and 5.8, J_g = 10.8 (H-2′b); 3.64 ddd, 1H, J(3′a,4′) = 3.9, J(3′a,P) = 7.0, J_g = 9.7 (H-3′a); 3.24 td, 1H, J(3′b,P) = 5.8, J(3′b,4′) = J_g = 9.8 (H-3′b); 1.82 m, 1H, J(4′,3′) = 3.9 and 10.0, J(4′,P) = J(4′,5′) = 7.1 (H-4′); 0.93 dd, 3H, J(5′,4′) = 7.1, J(5′,P) = 17.3 (H-5′). ^{13}C NMR (DMSO-d_6): 156.91 (C-6); 148.65 (C-4); 145.68 (C-2); 140.93 (C-8); 124.01 (C-5); 71.10 d, J(P,C) = 3.6 (C-3′); 68.33 (C-2′); 43.41 (C-1′); 31.12 d, J(P,C) = 135.6 (C-4′); 11.91 d, J(P,C) = 5.1 (C-5′). MS (ESI): m/z = 301 $[M-H]^-$. Anal. Calcd for $C_{10}H_{15}N_4O_5P\cdot1/2$MeOH: C, 39.69; H, 5.23; N, 17.63. Found: C, 39.62; H, 5.32; N, 17.27.

5.14.2. 9-[2-(2-Phosphonobutoxy)ethyl]hypoxanthine (19b)

Starting from bromo derivative **17b**, colorless foam, yield 68%. ^{1}H NMR (DMSO-d_6): 12.26 br s (OH); 8.06 s, 1H and 8.03 s, 1H (H-2, H-8); 4.30 t, 2H, J(1′,2′) = 5.3 (H-1′); 3.75 dt, 1H, and 3.70 dt, 1H, J(2′,1′) = 5.3, J_g = 10.6 (H-2′); 3.63 ddd, 1H, J(3′a,4′) = 3.9, J_g = 9.7, J(3′a,P) = 10.2 (H-3′a); 3.40 ddd, 1H, J(3′b,4′) = 8.9, J_g = 9.7, J(3′b,P) = 6.7 (H-3′b); 1.65 m, 1H (H-4′); 1.45 m, 2H (H-5′); 0.80 t, 3H, J(6′,5′) = 7.5 (H-6′). ^{13}C NMR (DMSO-d_6): 157.07 (C-6); 148.83 (C-4); 145.88 (C-2); 141.12 (C-8); 124.11 (C-5); 69.70 d, J(P,C) = 2.7 (C-3′); 68.66 (C-2′); 43.65 (C-1′); 39.97 d, J(P,C) = 134.0 (C-4′); 19.98 d, J(P,C) = 3.8 (C-5′); 12.55 d, J(P,C) = 7.6 (C-6′). MS (ESI): m/z = 315 $[M-H]^-$. Anal. Calcd for $C_{11}H_{17}N_4O_5P\cdot1/2H_2O$: C, 40.62; H, 5.58; N, 17.23. Found: C, 40.50; H, 5.42; N, 16.85.

5.14.3. 9-[2-(3-Phenyl-2-phosphonopropoxy)ethyl]hypoxanthine (19c)

Starting from bromo derivative **17c**, white solid, yield 60%. ^{1}H NMR (DMSO-d_6): 12.29 br s, 1H (OH); 8.03 s, 1H and 8.01 s, 1H (H-2, H-8); 7.14 m, 3H and 6.97 m, 2H (Ar); 4.25 br t, 2H, J(1′,2′) = 4.8 and 5.6 (H-1′); 3.62 ddd, 1H, and 3.58 ddd, J(2′,1′) = 4.8 and 5.6, J_g = 10.3 (H-2′); 3.40 m, 2H (H-3′); 2.87 td, 1H, J(5′a,4′) = 4.8, J(5′,P) = J_g = 13.8 (H-5′a); 2.61 ddd, 1H, J(5′b,4′) = 9.1, J(5′,P) = 9.4, J_g = 13.8 (H-5′b); 2,00 m, 1H (H-4′). ^{13}C NMR (DMSO-d_6): 156.82 (C-6); 148.48 (C-4); 145.59 (C-2); 140.98 (C-8); 140.31 d, J(P,C) = 11.8 (Ar); 129.02, 2C (Ar); 128.12, 2C (Ar); 125.93 (Ar); 123.89 (C-5); 68.54 br s (C-3′); 68.43 (C-2′); 43.35 (C-1′); 41.11 d, J(P,C) = 134.6 (C-4′); 32.17 d, J(P,C) = 2.0 (C-5′). MS (ESI): m/z = 377 $[M-H]^-$. Anal. Calcd for $C_{16}H_{19}N_4O_5P\cdot3/4H_2O$: C, 49.04; H, 5.27; N, 14.30. Found: C, 49.12; H, 5.07; N, 13.91.

5.14.4. 9-[2-(1-Phosphonopropan-2-yloxy)ethyl]hypoxanthine (31a)

Starting from bromo derivative **29a**, colorless foam, yield 90%. ^{1}H NMR (DMSO-d_6): 12.27 br s, 1H (NH); 8.05 s, 1H and 8.03 s, 1H (H-2 and H-8); 4.25 t, 2H, J(1′,2′) = 5.4 (H-1′); 3.72 t, 2H, J(2′,1′) = 5.4 (H-2′); 3.64 m, 1H (H-3′); 1.86 ddd, 1H, J(4′a,3′) = 4.1, J_g = 14.7, J(4′a,P) = 19.8 (H-4′a); 1.58 ddd, 1H, J(4′b,3′) = 8.8, J_g = 14.7, J(4′b,P) = 17.4 (H-4′b); 1.13 d, 3H, J(5′,3′) = 6.1 (H-5′). ^{13}C NMR (DMSO-d_6): 156.63 (C-6); 148.32 (C-4); 145.38 (C-2); 140.69 (C-8); 123.71 (C-5); 71.41 (C-3′); 65.47 (C-2′); 43.51 (C-1′); 35.26 d, J(P,C) = 132.3 (C-4′); 20.84 d, J(P,C) = 4.7 (C-5′). MS (ESI): m/z = 301 $[M-H]^-$. Anal. Calcd for $C_{10}H_{15}N_4O_5P\cdot2/3H_2O$: C, 38.22; H, 5.24; N, 17.83. Found: C, 38.34; H, 5.15; N, 17.52.

5.14.5. 9-[2-(1-Phosphonobutan-2-yloxy)ethyl]hypoxanthine (31b)

Starting from bromo derivative **29b**, colorless foam, yield 84%. ^{1}H NMR (DMSO-d_6): 12.32 br s, 1H (NH); 8.11 s, 1H and 8.04 s, 1H (H-2 and H-8); 4.28 t, 2H, J(1′,2′) = 5.3 (H-1′); 3.82 dt, 1H, J(2′a,1′) = 4.8, J_g = 10.5 (H-2′a); 3.67 dt, 1H, J(2′b,1′) = 5.6, J_g = 10.5 (H-2′b); 3.46 m, 1H (H-3′); 1.79 ddd, 1H, J(4′a,3′) = 4.7, J_g = 14.9, J(4′a,P) = 19.6 (H-4′a); 1.60 ddd, 1H, J(4′b,3′) = 7.9, J_g = 14.9, J(4′b,P) = 17.6 (H-4′b); 1.60 m, 1H, J(5′a,3′) = 4.0, J(5′a,6′) = 7.3, J_g = 14.2 (H-5′a); 1.37 sept, 1H, J(5′b,3′) = J(5′b,6′) = 7.2, J_g = 14.2 (H-5′b); 0.65 t, 3H, J(6′,5′) = 7.3 (H-6′). ^{13}C NMR (DMSO-d_6): 156.69 (C-6); 148.49 (C-4); 145.70 (C-2); 140.91 (C-8); 123.61 (C-5); 76.39 (C-3′); 66.06 (C-2′); 43.91 (C-1′); 32.16 d, J(P,C) = 132.4 (C-4′); 27.23 d, J(P,C) = 5.8 (C-5′); 8.99 (C-6′). MS (ESI): m/z = 315 $[M-H]^-$. Anal. Calcd for $C_{11}H_{17}N_4O_5P\cdot3/4H_2O$: C, 40.06; H, 5.65; N, 16.99. Found: C, 40.06; H, 5.50; N, 16.73.

5.14.6. 9-[2-(3-Phenyl-1-phosphonopropan-2-yloxy)ethyl]hypoxanthine (31c)

Starting from bromo derivative **29c**, colorless foam, yield 54%. ^{1}H NMR (DMSO-d_6): 12.25 br s, 1H (OH); 7.99 s, 1H and 7.91 s, 1H (H-2 and H-8); 7.12 m, 3H and 7.00 m, 2H (Ar); 4.22 ddd, 1H, J(1′,2′) = 4.4 and 7.0, J_g = 14.4 (H-1′a); 4.16 dt, 1H, J(1′,2′) = 4.5, J_g = 14.4 (H-1′b); 3.80 br dt, 1H, J(2′,1′) = 4.5, J_g = 10.4 (H-2′a); 3.59 ddd, 1H, J(2′,1′) = 7.0 and 4.5, J_g = 10.4 (H-2′b); 3.75 m, 1H (H-3′); 2.91 dd, 1H, J(5′a,3′) = 4.1, J_g = 13.9 (H-5′a); 2.65 dd, 1H, J(5′b,3′) = 7.1, J_g = 13.9 (H-5′b); 1.79 ddd, 1H, J(4′a,3′) = 5.2, J_g = 14.9, J(4′a,P) = 19.1 (H-4′a); 1.65 ddd, 1H, J(4′b,3′) = 7.5,J_g = 14.9, J(4′b,P) = 17.5 (H-4′b). ^{13}C NMR (DMSO-d_6): 156.58 (C-6); 148.18

(C-4); 145.16 (C-2); 140.53 (C-8); 138.26 (Ar); 129.22, 2C (Ar); 127.74, 2C (Ar); 125.75 (Ar); 123.73 (C-5); 76.26 (C-3′); 66.29 (C-2′); 43.50 (C-1′); 40.36 d, J(P,C) = 7.3 (C-5′); 32.50 d, J(P,C) = 132.5 (C-4′). MS (ESI): m/z = 377 $[M-H]^-$. Anal. Calcd for $C_{16}H_{19}N_4O_5P\cdot H_2O$: C, 48.49; H, 5.34; N, 14.14. Found: C, 48.44; H, 5.00; N, 14.26.

5.15. Synthesis of 9-[2-(phosphonoalkoxy)ethyl]guanines 20 and 32—general procedure

A mixture of 2-amino-6-bromoderivative **18** or **30** (2 mmol) was dissolved in trifluoroacetic acid (75%, 20 ml) and stirred overnight at room temperature. After evaporation and codistillation with water, the residue was applied onto a column of Dowex 50 × 8 (H^+-form, 40 ml) and washed with water. Elution with 2.5% aqueous ammonia and evaporation afforded crude product as ammonium salt. This residue was applied onto a column of Dowex 1 × 2 (acetate, 50 ml), washed with water followed by gradient of formic acid (0–0.5 M). The main UV-absorbing fraction was evaporated and codistilled with water to obtain product as a white solid.

5.15.1. 9-[2-(2-Phosphonopropoxy)ethyl]guanine (20a)

Starting from 2-amino-6-bromoderivative **18a**, yield 77%. ^{1}H NMR (DMSO-d_6): 10.70 br s, 1H (OH); 7.87 s, 1H (H-8); 6.55 br s, 2H (NH_2); 4.11 t, 2H, J(1′,2′) = 5.4 (H-1′); 3.77 dt, 1H, and 3.63 dt, 1H, J(2′,1′) = 5.4, J_g = 10.6 (H-2′); 3.65 ddd, 1H, J(3′a,4′) = 3.9, J(3′a,P) = 7.3, J_g = 9.6 (H-3′a); 3.26 td, 1H, J(3′b,P) = 6.0, J(3′b,4′) = 10.0, J_g = 9.6 (H-3′b); 1.85 m, 1H, J(4′,3′) = 3.9 and 10.0, J(4′,P) = J(4′,5′) = 7.1 (H-4′); 0.98 dd, 3H, J(5′,4′) = 7.1, J(5′,P) = 17.3 (H-5′). ^{13}C NMR (DMSO-d_6): 156.20 (C-6); 153.70 (C-2); 150.86 (C-4); 137.69 (C-8); 118.68 (C-5); 70.73 d, J(P,C) = 3.6 (C-3′); 67.81 (C-2′); 42.64 (C-1′); 32.78 d, J(P,C) = 135.3 (C-4′); 11.64 d, J(P,C) = 4.8 (C-5′). MS (ESI): m/z = 316 $[M-H]^-$. Anal. Calcd for $C_{10}H_{16}N_5O_5P\cdot 5/4H_2O$: C, 35.35; H, 5.49; N, 20.61. Found: C, 35.35; H, 5.31; N, 20.71.

5.15.2. 9-[2-(2-Phosphonobutoxy)ethyl]guanine (20b)

Starting from 2-amino-6-bromoderivative **18b**, yield 56%. ^{1}H NMR (DMSO-d_6): 10.53 br s, 1H (OH); 7.65 s, 1H (H-8); 6.43 br s, 2H (NH_2); 4.08 t, 2H, J(1′,2′) = 5.3 (H-1′); 3.65 m, 3H, (H-2′ and H-3′a); 3.38 td, 1H, J(3′b,4′) = J_g = 9.4, J(3′b,P) = 6.5 (H-3′b); 1.67 m, 1H (H-4′); 1.49 m, 2H (H-5′); 0.86 t, 3H, J(6′,5′) = 7.5 (H-6′). ^{13}C NMR (DMSO-d_6): 156.95 (C-6); 153.67 (C-2); 151.32 (C-4); 137.98 (C-8); 116.47 (C-5); 69.49 d, J(P,C) = 2.8 (C-3′); 68.39 (C-2′); 42.68 (C-1′); 39.90 d, J(P,C) = 134.9 (C-4′); 19.76 d, J(P,C) = 3.8 (C-5′); 12.34 d, J(P,C) = 7.6 (C-6′). MS (ESI): m/z = 330 $[M-H]^-$. Anal. Calcd for $C_{11}H_{18}N_5O_5P\cdot 1/4H_2O$: C, 39.35; H, 5.55; N, 20.86. Found: C, 39.45; H, 5.41; N, 20.75.

5.15.3. 9-[2-(3-Phenyl-2-phosphonopropoxy)ethyl]guanine (20c)

Starting from 2-amino-6-bromoderivative **18c**, yield 81%. ^{1}H NMR (DMSO-d_6): 10.57 br s, 1H (OH); 7.63 s, 1H (H-8); 6.45 br s, 2H (NH_2); 7.16 m, 3H and 7.00 m, 2H (Ar); 4.04 br t, 2H, J(1′,2′) = 4.9 and 5.4 (H-1′); 3.55 d br t, 1H, and 3.50 d br t, J(2′,1′) = 4.9 and 5.4, J_g = 10.3 (H-2′); 3.40 m, 2H (H-3′); 2.88 td, 1H, J(5′a,4′) = 4.8, J(5′,P) = J_g = 13.9 (H-5′a); 2.62 ddd, 1H, J(5′b,4′) = 9.0, J(5′,P) = 10.2, J_g = 13.9 (H-5′b); 1.99 m, 1H (H-4′). ^{13}C NMR (DMSO-d_6): 157.22 (C-6); 154.00 (C-2); 151.53 (C-4); 138.47 (C-8); 140.66 d, J(P,C) = 11.8 (Ar); 129.40, 2C (Ar); 128.44, 2C (Ar); 126.22 (Ar); 116.67 (C-5); 68.84 br s (C-3′); 68.67 (C-2′); 43.02 (C-1′); 41.72 d, J(P,C) = 135.0 (C-4′); 32.50 br s (C-5′). MS (ESI): m/z = 392 $[M-H]^-$. Anal. Calcd for $C_{16}H_{20}N_5O_5P\cdot 4/5H_2O$: C, 47.13; H, 5.34; N, 17.18. Found: C, 47.16; H, 5.35; N, 16.88.

5.15.4. 9-[2-(1-Phosphonopropan-2-yloxy)ethyl]guanine (32a)

Starting from 2-amino-6-bromoderivative **30a**, yield 80%. ^{1}H NMR (DMSO-d_6): 10.55 br s, 1H (OH); 7.66 s, 1H (H-8); 6.44 br s, 2H (NH_2); 4.04 t, 2H, J(1′,2′) = 5.5 (H-1′); 3.64 m, 3H (H-2′ and H-3′); 1.88 ddd, 1H, J(4′a,3′) = 4.2, J_g = 14.8, J(4′a,P) = 19.7 (H-4′a); 1.57 ddd, 1H, J(4′b,3′) = 8.8, J_g = 14.8, J(4′b,P) = 17.4 (H-4′b), 1.55 d, 3H, J(5′,3′) = 6.1 (H-5′). ^{13}C NMR (DMSO-d_6): 156.75 (C-6); 153.48 (C-2); 151.09 (C-4); 137.83 (C-8); 116.24 (C-5); 71.42 (C-3′); 65.49 (C-2′); 42.82 (C-1′); 35.34 d, J(P,C) = 132.4 (C-4′); 21.02 d, J(P,C) = 4.6 (C-5′). MS (ESI): m/z = 316 $[M-H]^-$. Anal. Calcd for $C_{10}H_{16}N_5O_5P\cdot 3/4H_2O$: C, 36.31; H, 5.33; N, 21.17. Found: C, 36.41; H, 5.23; N, 20.86.

5.15.5. 9-[2-(1-Phosphonobutan-2-yloxy)ethyl]guanine (32b)

Starting from 2-amino-6-bromoderivative **30b**, yield 67%. ^{1}H NMR (DMSO-d_6): 10.68 br s, 1H (OH); 7.86 s, 1H (H-8); 6.53 br s, 2H (NH_2); 4.08 t, 2H, J(1′,2′) = 5.6 (H-1′); 3.75 dt, 1H, J(2′a,1′) = 5.1, J_g = 10.4 (H-2′a), 3.61 dt, 1H, J(2′b,1′) = 6.2, J_g = 10.4 (H-2′b), 3.48 m, 2H (H-3′); 1.81 ddd, 1H, J(4′a,3′) = 4.7, J_g = 14.9, J(4′a,P) = 19.6 (H-4′a); 1.64 ddd, 1H, J(4′b,3′) = 7.8, J_g = 14.9, J(4′b,P) = 17.6 (H-4′b); 1.64 m, 1H, J(5′a,3′) =4.0, J(5′a,6′) = 7.3, J_g = 14.2 (H-5′b), 1.40 sept, 1H, J(5′b,3′) = J(5′b,6′) = 7.2, J_g = 14.2 (H-5′b), 0.70, 3H, J(6′,5′) = 7.3 (H-6′). ^{13}C NMR (DMSO-d_6): 156.53 (C-6); 153.89 (C-2); 151.11 (C-4); 138.03 (C-8); 115.31 (C-5); 76.35 (C-3′); 65.98 (C-2′); 43.36 (C-1′); 32.16 d, J(P,C) = 132.9 (C-4′); 27.23 d, J(P,C) = 6.0 (C-5′); 9.03 (C-6′).MS (ESI): m/z = 330 $[M-H]^-$. Anal. Calcd for $C_{11}H_{18}N_5O_5P\cdot 1/5H_2O$: C, 39.45; H, 5.54; N, 20.91. Found: C, 39.63; H, 5.46; N, 20.65.

5.15.6. 9-[2-(3-Phenyl-1-phosphonopropan-2-yloxy)ethyl]guanine (32c)

Starting from 2-amino-6-bromoderivative **30c**, yield 68%. ^{1}H NMR (DMSO-d_6): 10.64 br s, 1H (OH); 7.55 s, 1H (H-8); 7.16 m, 3H and 7.08 m, 2H (Ar); 6.43 br s, 2H (NH_2); 4.03 ddd, 1H, J(1′,2′) = 4.4 and 7.1, J_g = 14.2 (H-1′a); 3.96 dt, 1H, J(1′,2′) = 4.7, J_g = 14.2 (H-1′b); 3.71 br dt, 1H, J(2′,1′) = 4.6, J_g = 10.5 (H-2′a); 3.55 ddd, 1H, J(2′,1′) = 7.1 and 4.5, J_g = 10.5 (H-2′b); 3.77 m, 1H (H-3′); 2.94 dd, 1H, J(5′a,3′) = 4.4, J_g = 13.8 (H-5′a); 2.68 dd, 1H, J(5′b,3′) = 7.0, J_g = 13.8 (H-5′b); 1.79 ddd, 1H, J(4′a,3′) = 5.1, J_g = 14.9, J(4′a,P) = 19.1 (H-4′a); 1.66 ddd, 1H, J(4′b,3′) = 7.5, J_g = 14.9, J(4′b,P) = 17.3 (H-4′b). ^{13}C NMR (DMSO-d_6): 156.95 (C-6); 153.62 (C-2); 151.23 (C-4); 138.57 (Ar); 137.98 (C-8); 129.63, 2C (Ar); 128.09, 2C (Ar); 126.08 (Ar); 116.47 (C-5); 76.51 (C-3′); 66.60 (C-2′); 43.09 (C-1′); 40.54 d, J(P,C) = 7.4 (C-5′); 32.75 d, J(P,C) = 133.5 (C-4′). MS (ESI): m/z = 392 $[M-H]^-$. Anal. Calcd for $C_{16}H_{20}N_5O_5P\cdot 7/8H_2O$: C, 45.23; H, 5.58; N, 16.48. Found: C, 45.08; H, 5.40; N, 16.27.

5.16. Synthesis of 9-[2-(phosphonoalkoxy)ethyl]xanthines 21 and 33—general procedure

A mixture of guanine derivative **20** or **32** (1 mmol), aqueous HCl (1 M, 30 ml) and $NaNO_2$ (0.3 g, 4.3 mmol) was stirred for 30 min at room temperature and then a solution of NaOH (1 M) was added to pH 9. The reaction mixture was diluted with water (200 ml) and applied onto a column of Dowex 1 × 2 (acetate, 100 ml). The column was washed with water followed by gradient of formic acid (0.1–1 M). The main UV-absorbing fraction was evaporated and codistilled with water (3×). Pure product was obtained after preparative HPLC as a white amorphous solid.

5.16.1. 9-[2-(2-Phosphonopropoxy)ethyl]xanthine (21a)

Starting from guanine derivative **20a**, yield 76%. ^{1}H NMR (DMSO-d_6): 11.83 br s, 1H and 10.77 s (OH–NH); 7.63 s, 1H (H-8); 4.19 m, 2H (H-1′); 3.65 ddd, 1H, and 3.58 ddd, 1H, J(2′,1′) = 4.5 and 5.6, J_g = 10.7 (H-2′); 3.60 m, 1H (H-3′a); 3.25 td,

1H, J(3′b,4′) = 5.7, J_g = J(3′b,P) = 9.9 (H-3′b); 1.83 m, 1H (H-4′); 1.00 dd, 3H, J(5′,4′) = 7.0, J(5′,P) = 17.4 (H-6′). ^{13}C NMR (DMSO-d_6): 157.79 (C-6); 150.66 (C-2); 140.37 (C-4); 137.20 (C-8); 115.07 (C-5); 71.12 d, J(P,C) = 3.9 (C-3′); 68.14 (C-2′); 43.74 (C-1′); 32.76 d, J(P,C) = 135.3 (C-4′); 11.45 d, J(P,C) = 5.3 (C-5′). MS (ESI): m/z = 313 [M−H]$^-$. Anal. Calcd for $C_{10}H_{15}N_4O_6P\cdot 3/5MeOH$: C, 37.73; H, 5.20; N, 16.60. Found: C, 38.05; H, 5.13; N, 16.34.

5.16.2. 9-[2-(2-Phosphonobutoxy)ethyl]xanthine (21b)

Starting from guanine derivative **20b**, yield 59%. ^{1}H NMR (DMSO-d_6): 11.80 br s, 1H and 10.76 s (OH-NH); 7.62 s, 1H (H-8); 4.19 t, 2H, J(1′,2′) = 5.3 (H-1′); 3.76 dt, 1H, and 3.68 dt, 1H, J(2′,1′) = 5.3, J_g = 10.8 (H-2′); 3.73 ddd, 1H, J(3′a,4′) = 3.8, J_g = 9.7, J(3′a,P) = 10.5 (H-3′a); 3.37 ddd, 1H, J(3′b,4′) = 9.1, J_g = 9.7, J(3′b,P) = 6.4 (H-3′b); 1.65 m, 1H (H-4′); 1.47 m, 2H (H-5′); 0.82 t, 3H, J(6′,5′) = 7.5 (H-6′). ^{13}C NMR (DMSO-d_6): 158.41 (C-6); 151.27 (C-2); 141.01 (C-4); 137.76 (C-8); 115.70 (C-5); 69.86 d, J(P,C) = 2.9 (C-3′); 68.93 (C-2′); 44.36 (C-1′); 40.19 d, J(P,C) = 134.4 (C-4′); 19.98 d, J(P,C) = 3.7 (C-5′); 12.56 d, J(P,C) = 7.5 (C-6′). MS (ESI): m/z = 331 [M−H]$^-$. Anal. Calcd for $C_{11}H_{17}N_4O_6P\cdot 1/2H_2O$: C, 38.71; H, 5.32; N, 16.42. Found: C, 38.86; H, 5.26; N, 16.53.

5.16.3. 9-[2-(3-Phenyl-2-phosphonopropoxy)ethyl]xanthine (21c)

Starting from guanine derivative **20c**, yield 72%. ^{1}H NMR (DMSO-d_6): 11.84 br s, 1H and 10.80 s (OH–NH); 7.57 s, 1H (H-8); 7.16 m, 3H and 7.00 m, 2H (Ar); 4.19 br t, 2H, J(1′,2′) = 4.9 and 5.2 (H-1′); 3.55 m, 4H (H-2′ and H-3′); 2.09 m, 1H (H-4′); 2.87 td, 1H, J(5′a,4′) = 5.0, J_g = J(5′a,P) = 13.8 (H-5′a); 2.61 ddd, 1H, J(5′b,4′) = 8.6, J_g = 13.9, J(5′a,P) = 10.5 (H-5′b). ^{13}C NMR (DMSO-d_6): 157.87 (C-6); 150.74 (C-2); 140.31 (C-4); 140.02 d, J(P,C) = 12.0 (Ar); 137.40 (C-8); 128.79, 2C, 127.86, 2C and 125.69 (Ar);115.20 (C-5); 68.41 (C-3′); 68.30 (C-2′); 43.88 (C-1′); 40.20 d, J(P,C) = 134.3 (C-4′); 31.88 d, J(P,C) = 1.5 (C-5′). MS (ESI): m/z = 393 [M−H]$^-$. Anal. Calcd for $C_{16}H_{19}N_4O_6P\cdot 2/3H_2O$: C, 47.29; H, 5.04; N, 13.79. Found: C, 47.42; H, 4.95; N, 13.66.

5.16.4. 9-[2-(1-Phosphonopropan-2-yloxy)ethyl]xanthine (33a)

Starting from guanine derivative **32a**, yield 53%. ^{1}H NMR (DMSO-d_6): 11.90 br s, 1H and 10.76 s, 1H (NH, OH); 7.63 s, 1H (H-8); 4.15 t, 2H, J(1′,2′) = 5.2 (H-1′); 3.61 t, 2H, J(2′,1′) = 5.2 (H-2′); 3.64 m, 1H (H-3′); 1.86 ddd, 1H, J(4′a,3′) = 4.3, J_g = 14.7, J(4′a,P) = 19.6 (H-4′a); 1.57 ddd, 1H, J(4′b,3′) = 8.6, J_g = 14.7, J(4′b,P) = 17.5 (H-4′b); 1.13 d, 3H, J(5′,3′) = 6.1 (H-5′). ^{13}C NMR (DMSO-d_6): 157.85 (C-6); 150.70 (C-2); 140.42 (C-4); 137.27 (C-8); 115.13 (C-5); 71.58 (C-3′); 65.76 (C-2′); 44.17 (C-1′); 35.11 d, J(P,C) = 132.4 (C-4′); 20.87 d, J(P,C) = 5.2 (C-5′). MS (ESI): m/z = 317 [M−H]$^-$. Anal. Calcd for $C_{10}H_{15}N_4O_6P\cdot 2/3H_2O$: C, 36.37; H, 4.99; N, 16.97. Found: C, 36.29; H, 4.79; N, 16.64.

5.16.5. 9-[2-(1-Phosphonobutan-2-yloxy)ethyl]xanthine (33b)

Starting from guanine derivative **32b**, yield 60%. ^{1}H NMR (DMSO-d_6): 11.90 br s, 1H and 10.77 s, 1H (NH, OH); 7.63 s, 1H (H-8); 4.28 t, 2H, J(1′,2′) = 5.3 (H-1′); 3.82 dt, 1H, J(2′a,1′) = 4.7, J_g = 10.6, (H-2′a); 3.66 dt, 1H, J(2′b,1′) = 6.1, J_g = 10.6, (H-2′b); 3.57 m, 1H (H-3′); 1.91 ddd, 1H, J(4′a,3′) = 4.9, J_g = 15.0, J(4′a,P) = 19.5 (H-4′a); 1.72 m, 2H (H-4′b and H-5′a); 1.49 sept, 1H, J(5′b,3′) = J(5′b,6′) = 7.2, J_g= 14.2 (H-5′b); 0.79 t, 3H, J(6′,5′) = 7.3 (H-6′). ^{13}C NMR (DMSO-d_6): 158.13 (C-6); 150.98 (C-2); 140.75 (C-4); 137.53 (C-8); 115.41 (C-5); 76.65 (C-3′); 66.67 (C-2′); 44.51 (C-1′); 32.17 d, J(P,C) = 132.8 (C-4′); 27.47 d, J(P,C) = 6.1 (C-5′); 9.02 (C-6′). MS (ESI): m/z = 331 [M−H]$^-$. Anal. Calcd for $C_{11}H_{17}N_4O_6P\cdot 1/2H_2O$: C, 38.71; H, 5.32; N, 16.42. Found: C, 38.70; H, 5.28; N, 16.15.

5.16.6. 9-[2-(3-Phenyl-1-phosphonopropan-2-yloxy)ethyl]xanthine (33c)

Starting from guanine derivative **32c**, yield 71%. ^{1}H NMR (DMSO-d_6): 11.75 br s, 1H and 10.75 s, 1H (NH/OH); 7.53 s, 1H (H-8); 7.14 m, 3H and 7.03 m, 2H (Ar); 4.13 ddd, 1H, J(1′,2′) = 4.0 and 7.3, J_g = 14.6 (H-1′a); 4.07 dt, 1H, J(1′,2′) = 4.4, J_g = 14.6 (H-1′b); 3.68 br dt, 1H, J(2′,1′) = 4.0, J_g = 10.5 (H-2′a); 3.46 ddd, 1H, J(2′,1′) = 7.3 and 3.7, J_g = 10.5 (H-2′b); 3.77 m, 1H (H-3′); 2.92 dd, 1H, J(5′a,3′) = 4.2, J_g = 13.9 (H-5′a); 2.66 dd, 1H, J(5′b,3′) = 7.0, J_g = 13.9 (H-5′b); 1.81 ddd, 1H, J(4′a,3′) = 5.7, J_g = 15.0, J(4′a,P) = 19.0 (H-4′a); 1.67 ddd, 1H, J(4′b,3′) = 7.0, J_g = 15.0, J(4′b,P) = 17.5 (H-4′b). ^{13}C NMR (DMSO-d_6): 158.16 (C-6); 150.95 (C-2); 140.62 (C-4); 138.48 (Ar); 137.43 (C-8); 129.48, 2C (Ar); 128.03, 2C (Ar); 126.05 (Ar); 115.52 (C-5); 76.74 (C-3′); 67.11 (C-2′); 44.48 (C-1′); 40.85 d, J(P,C) = 7.4 (C-5′); 32.76 d, J(P,C) = 133.2 (C-4′). MS (ESI): m/z = 393 [M−H]$^-$. Anal. Calcd for $C_{16}H_{19}N_4O_6P\cdot H_2O$: C, 46.61; H, 5.13; N, 13.59. Found: C, 46.43; H, 5.05; N, 13.41.

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Supplementary data

Supplementary data (Synthesis of the compounds **3**, **4** and **6**) associated with this article can be found, in the online version, at doi:10.1016/j.bmc.2009.07.044.

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