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Synthesis of 9-phosphonoalkyl and 9-phosphonoalkoxyalkyl purines: Evaluation of their ability to act as inhibitors of *Plasmodium falciparum*, *Plasmodium vivax* and human hypoxanthine–guanine–(xanthine) phosphoribosyltransferases

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ABSTRACT

The purine salvage enzyme, hypoxanthine–guanine–(xanthine) phosphoribosyltransferase [HG(X)PRT], catalyses the synthesis of the purine nucleoside monophosphates, IMP, GMP or XMP essential for DNA/RNA production. In protozoan parasites, such as *Plasmodium*, this is the only route available for their synthesis as they lack the de novo pathway which is present in human cells. Acyclic nucleoside phosphonates (ANPs), analogs of the purine nucleoside monophosphates, have been found to inhibit *Plasmodium falciparum* (*Pf*) HGXPRT and *Plasmodium vivax* (*Pv*) HGPRT with K_i values as low as 100 nM. They arrest parasitemia in cell based assays with IC_{50} values of the order of 1–10 μM. ANPs with phosphonoalkyl and phosphonoalkoxyalkyl moieties linking the purine base and phosphonate group were designed and synthesised to evaluate the influence of this linker on the potency and/or selectivity of the ANPs for the human and malarial enzymes. This data shows that variability in the linker, as well as the positioning of the oxygen in this linker, influences binding. The human enzyme binds the ANPs with K_i values of 0.5 μM when the number of atoms in the linker was 5 or 6 atoms. However, the parasite enzymes have little affinity for such long chains unless oxygen is included in the three-position. In comparison, all three enzymes have little affinity for ANPs where the number of atoms linking the base and the phosphonate group is of the order of 2–3 atoms. The chemical nature of the purine base also effects the K_i values. This data shows that both the linker and the purine base play an important role in the binding of the ANPs to these three enzymes.

1. Introduction

Malaria remains one of the most serious infectious diseases in the world today. Due to increasing resistance to current medications, there is a need to develop new classes of antimalarial drugs.[1] *Plasmodium falciparum* (*Pf*) and *Plasmodium vivax* (*Pv*) are the most widespread species that cause malaria in humans. *Pf* is reputed to be the most lethal but *Pv* is also responsible for serious illness with recurring bouts of fever.[2]

One significant difference in the metabolic pathways between *Plasmodium* and its human host cell is in the ability to synthesise the purine nucleoside monophosphates essential for the production of DNA/RNA. Mammalian cells are able to produce these metabolites either by de novo synthesis or by salvage. In contrast, the malarial parasite possesses only one pathway and this is the salvage of preformed bases transported from its host cell. Hypoxanthine–guanine–xanthine phosphoribosyltransferase (HGXPRT) is the purine salvage enzyme which catalyses the reaction shown in Fig. 1.[3,4]

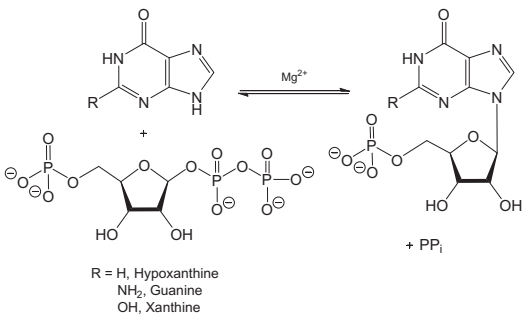

Figure 1. Reaction catalyzed by HGXPRT.

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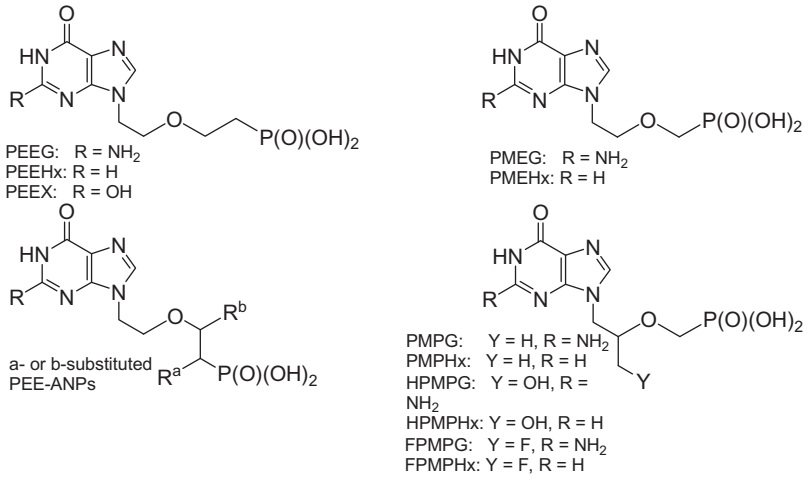


Figure 2. ANP inhibitors of HG(X)PRT with linear and branched chains.

Thus, this enzyme plays a crucial role in the replication and survival of the parasite and is therefore a target for the design of antiparasitic drugs.

Acyclic nucleoside phosphonates (ANPs) are inhibitors of *Pf*HGXPRT and have been found to arrest parasitemia in cells grown in culture.[5] These compounds are structural analogs of nucleoside monophosphates where the naturally occurring labile phosphate moiety is replaced by the phosphonate group containing a stable P–C bond. Molecules belonging to this family are known for their favorable pharmacokinetic profiles and low toxicity.[6]

As a result of these features, several ANPs have become drugs (e.g., Truvada®, Vistide®, Hepsera®) that are now in clinical use for the treatment of viral infections (HIV, HBV, CMV).[7]

Previously, a series of ANPs has been evaluated as inhibitors of *Pf*HGXPRT[5,8], *Pv*HGPRT[9] and human HGPRT.

These ANPs can have linear moieties linking the purine base and phosphonate group [PME: 2-(phosphonylmethoxy)ethyl and PEE: 2-(phosphonylethoxy)ethyl] or be derivatives with branched linkers [HPMP: 3-hydroxy-2-(phosphonomethoxy)propyl, PMP: 2-(phosphonomethoxy)propyl, FPMP: 3-fluoro-2-(phosphonomethoxy) propyl, α- and β-branched PEE], Fig. 2.

Crystal structures of three ANPs in complex with human HGPRT have shown that the phosphonate group binds in the 5′-phosphate binding pocket of the natural substrate of the reaction (GMP or IMP) and the purine base is anchored in the purine base binding site.[5,8] However, these enzymes are flexible and it is known that the human enzyme undergoes structural change when the substrates bind and the products are released. In the human HGPRT in complex with the ANPs, there are interactions between the linker and the amino acid side chains or backbone atoms at the active site. However, the precise contribution of this linker in increasing potency and/or selectivity has not been evaluated.

To continue this structure–activity relationship study, a number of 9-substituted phosphonoalkyl and phosphonoalkoxyalkyl purines were synthesised. The rationale for this synthesis was to investigate: (i) the effect of the length of the linker between the phosphonate group and the purine base; (ii) the influence of the oxygen atom in the linker; and (iii) the identity of the purine base itself on the inhibition of both human HGPRT and *Pf*HGXPRT. Herein, we report the synthesis and the kinetic constants for a comprehensive series of ANP compounds.

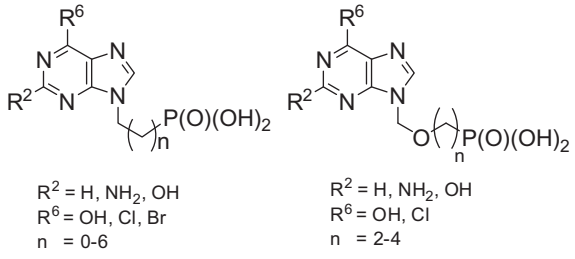


Figure 3. The structure of the 9-substituted phosphonoalkyl and phosphonoalkoxyalkyl purines.

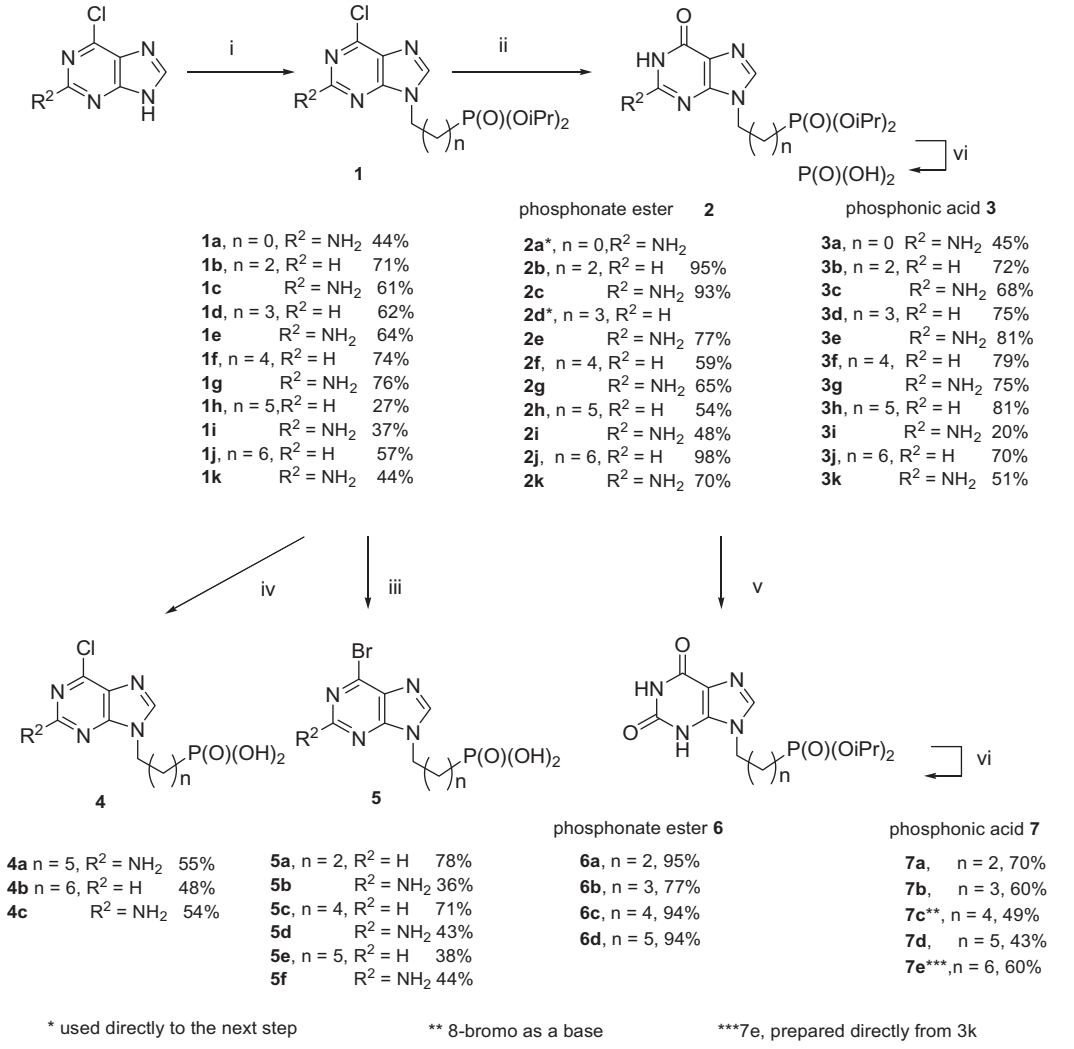
2. Results and discussion

2.1. Chemistry

The basic structure of the 9-substituted phosphonoalkyl and phosphonoalkoxyalkyl purines is shown in Fig. 3. The variability lies in the 2-position and 6-position of the purine base, the number of atoms between the base and the phosphonate group and the inclusion of oxygen in the tail.

Previous experience led us to prefer the classical reaction pathway with direct introduction of a suitable chain into the N^9-position of halogenopurines, followed by standard functional group transformations to prepare the corresponding guanine, xanthine and hypoxanthine derivatives.

6-Chloro and 2-amino-6-chloro derivatives **1a–k** and **9** were obtained by treatment of 6-chloropurine or 2-amino-6-chloropurine with appropriate halogenoalkyl phosphonates **8a**,[12] **8b**,**c**[10] **8d**,**e**[11] and **8f** in the presence of NaH (Scheme 1). The above mentioned halogenoalkyl phosphonates **8** were not prepared under conventional heating conditions, described in the original literature, but were subjected to an Arbuzov reaction under microwave conditions according to a recently improved procedure.[12] The most efficient ratio of dihalogenoalkane to triisopropylphosphite was 3:1. The obtained products were next used directly in the alkylation step. The alkylation by preformed halogenoalkyl phosphonates **8a–f** proceeded with the expected yields for $n = 0$ and 2–6, but the classical alkylation reaction by isopropyl bromoethylphosphonate[12] **8g** ($n = 1$) was inefficient. The isolated yield of **9** was very low (8%) probably due to an elimination reaction.[13] This problem was partially overcome by the reaction with diethyl vinylphosphonate under basic catalysis using caesium carbonate (Scheme 2). The free phosphonic acid **10** was afterwards prepared by the procedure described in literature.[14] Hypoxanthine derivatives **12a**



Scheme 1.

and **12b** were prepared by diazotation reaction[15] of the appropriate adenine congeners **11**[13] (Scheme 3).

In contrast with the above mentioned alkylation by haloalkyl phosphonates (Scheme 1), the enhanced reactivity of haloalkyloxyalkyl phosphonates **13a**[16] **13b** and **13c** (n = 2–4, prepared according to the literature[17]) allowed the use of mild alkylation conditions (−10 °C–rt) for the preparation of compounds **14a–f** (Scheme 4).

Two methods were used for the transformation of the 6-chloropurine derivatives **1** and **14** to the corresponding compounds with a 6-oxopurine base (Scheme 1 and 4): Compounds **1a**,**c**,**e**,**g**,**i** as well as **1b**,**f**,**h** were transformed to the corresponding guanine **2a**,**c**,**e**,**g**,**i** or hypoxanthine derivatives **2b**,**f**,**h**, respectively, by hydrolysis with 1 M HCl (Scheme 1).

Due to the instability of the α-oxo position in the side chain under acid conditions, basic hydrolysis was used to synthesise compounds **14a–d**,**f** (Scheme 4) and **1d**,**j**,**k**. The reflux of these compounds with DABCO and K_2CO_3[18] afforded the corresponding 6-oxopurine derivatives **15a–d**,**f** and **2j**,**k**. Compound **2d** was, under these conditions, partially transformed to the mono ethyl ester and both types of phosphonate esters were subsequently fully cleaved in the next step.

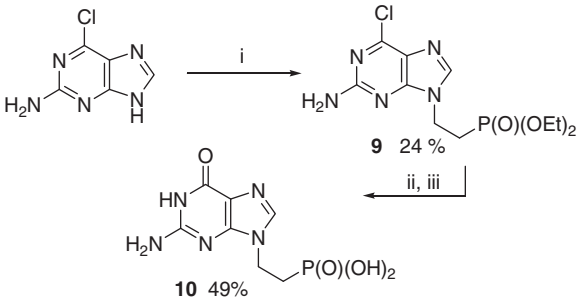
The cleavage of the phosphonate diesters **2a**,**c**,**e**,**g**,**i**,**k** by reaction with bromotrimethylsilane followed by hydrolysis afforded the free guanine derivatives **3a**,**c**,**e**,**g**,**i**,**k**. The previously known guanine derivatives **3e** and **3k** were thus prepared by the above mentioned alternative route instead of that reported previously.[19,20]

The same procedure was also used for the transformation of hypoxanthine diesters **2b**,**d**,**f**,**h**,**j** to the free phosphonates **3b**,**d**,**f**,**h**,**j**. The hypoxanthine derivative **3f** was thus prepared contrary to the literature[21] by a non-enzymatic procedure in the same way as the other hypoxanthine derivatives. Acid labile guanine and hypoxanthine derivatives **16a–f** were prepared by ester cleavage using TMSBr with addition of 2,6-lutidine.[8]

Protected guanines **2c**,**e**,**g**,**i**,**k** were also transformed to appropriate xanthine derivatives by the classical diazotation reaction. The obtained compounds **6a–d** were treated with TMSBr to afford xanthine ANPs **7a**,**b**,**d**. Compound **7c** was isolated as the 8-bromo derivative.

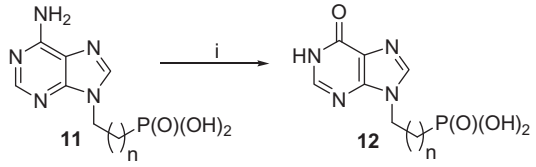
To eliminate this potential problem other xanthine compounds **7e** and **18a–c** were prepared directly from guanine phosphonic acids **3k** and **16b**,**d**,**f**, respectively.

The 6-chloro derivatives **1b–k** and **14a–f** were also transformed into free halogenopurine phosphonates. The presence of HBr as a



i) Diethyl vinylphosphonate, Cs_2CO_3, DMF, 90 °C;
ii) 1M HCl, reflux; iii) TMSBr, CH_3CN, DMF, RT

Scheme 2.



12a 35% **n = 0**
12b 43% **n = 1**

i) Amyl nitrite, 80% CH_3COOH

Scheme 3.

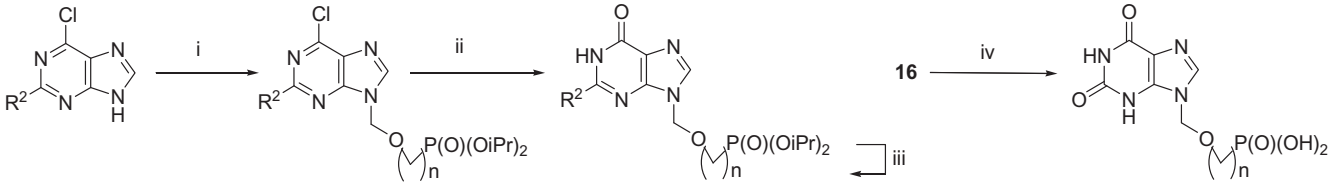
contaminant in TMSBr caused the substitution of the 6-chloro group by the bromine and the 6-bromoderivatives **5a–f** were isolated after ester cleavage of **1b–g**. To prevent this exchange of halogen, 2,6-lutidine was used to neutralize the HBr present and starting from **1h–k** and **14a–f** we isolated 6-chloropurine compounds **4a–c** and a series **17a–f**, respectively.

2.1.1. Inhibition of human HGPRT, *Pv*HGPRT and *Pf*HGXPRT by the alkyl and alkyloxyalkyl phosphonates

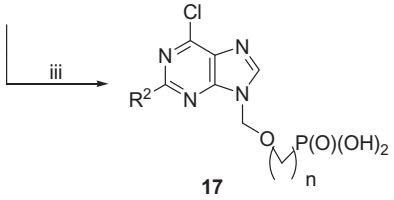
Table 1 gives the K_i values for the inhibition of human HGPRT, *Pf*HGXPRT and *Pv*HGPRT together with the description of structural variations of the ANPs.

In general, when the linker contains only one, two or three atoms, binding is virtually abolished. There are two exceptions to this rule which are compounds **12a** and **12b**. They have K_i values between 2 and 4 μM for human enzyme. **12a** does not bind to *Pf* HGXPRT or *Pv*HGPRT though **12b** has a K_i value of 2 μM. The underlying reason for the fact that the shorter compounds do not generally bind to the enzymes can be attributed to the fact that, if the base attempts to bind first, the phosphonate group cannot reach into the 5′-phosphate binding pocket or conversely if the phosphonate group manages to reach into the active site first, the base is too far away from its true binding site. It is not clear why compounds **12a** and **12b** can be inhibitors of these enzymes. Their positions in the active site may be stabilized by the presence of additional ordered water molecules that bridge the inhibitor to the enzyme. Alternatively, these inhibitors may be oriented in the active site in a completely different way as to what is observed when PEEG, other phosphonates or GMP (the product of the reaction) is bound.

Increasing the length of the linker to four atoms does result in binding, though weak, suggesting that the linker is now long enough to bridge the two binding sites which are the purine base and the 5′-phosphate binding pocket. The low K_i values (between 5–50 μM) may be due to the fact that binding of the purine base and the phosphonate group are not sufficient alone to ensure that the ANPs bind tightly. In contrast, PMEG which contains an oxygen in the 3-position in the linker binds reasonably well to *Pf*HGXPRT though, for human HGPRT, this change makes little difference in the affinity. The addition of an oxygen atom to the second or third position of the linker generally enhances binding for this set of compounds particularly for the *Plasmodium* enzymes. The addition



14a	n = 2, R^2 = H 32%	**15a**	n = 2, R^2 = H 47%	**16a**	n = 2, R^2 = H 61%	**18a**	n = 2, 57%
14b	R^2 = NH_2 38%	**15b**	R^2 = NH_2 91%	**16b**	R^2 = NH_2 54%	**18b**	n = 3, 48%
14c	n = 3, R^2 = H 56%	**15c**	n = 3, R^2 = H 49%	**16c**	n = 3, R^2 = H 86%	**18c**	n = 4, 58%
14d	R^2 = NH_2 61%	**15d**	R^2 = NH_2 69%	**16d**	R^2 = NH_2 58 %		
14e	n = 4, R^2 = H 37%	**15e**	n = 4,	**16e***	n = 4, R^2 = H 10%		
14f	R^2 = NH_2 40%	**15f**	R^2 = NH_2 65%	**16f**	R^2 = NH_2 56%		



17a	n = 2, R^2 = H 42%
17b	R^2 = NH_2 43%
17c	n = 3, R^2 = H 56%
17d	R^2 = NH_2 61%
17e	n = 4, R^2 = H 37%
17f	R^2 = NH_2 40%

* prepared from **17e**

i) Chlorophosphonates **13a-c**, NaH, DMF -10 °C - RT; ii) DABCO, K_2CO_3, H_2O, reflux;
iii) TMSBr, CH_3CN, 2,6-lutidine; iv) $NaNO_2$, 1M HCl, H_2O;

Scheme 4.

Table 1
Comparison of the K_i values for the alkyl and alkoxyalkyl phosphonates with human HGPRT, *Pv*HGPRT and P*f*HGXPRT.

Compound	Number of atoms[a]	Position of oxygen atom[b]	Base[e]	K_i (μM) Human	K_i (μM) *Pf*	K_i (μM) *Pv*
3a	1	NO[c]	G	>100	>100	>100
12a	1	NO[c]	Hx	1.9	>100	>100
10	2	NO[c]	G	>100	>100	>100
12b	2	NO[c]	Hx	3.4	2.2	>100
3c	3	NO[c]	G	>100	>100	>100
3b	3	NO[c]	Hx	>100	>100	>100
3e	4	NO[c]	G	21	>100	>100
3d	4	NO[c]	Hx	>100	>100	>100
16b	4	2	G	31	5	24
16a	4	2	Hx	56	25	30
PMEG[5]	4	3	G	29	1.6	ND[d]
3 g	5	NO[c]	G	0.5	2	6
3f	5	NO[c]	Hx	>100	21	ND[d]
16d	5	2	G	3	6.7	1
d	5	2	Hx	>100	47	>100
PEE[5]	5	3	G	1	0.1	ND[d]
PEEHx[5]	5	3	Hx	3.6	0.3	ND[d]
3i	6	NO[c]	G	0.5	>100	>100
3 h	6	NO[c]	Hx	77	97	ND
16f	6	2	G	0.4	200	>100
16e	6	2	Hx	28	29	ND[d]
3 k	7	NO[c]	G	5	10	ND[d]
3j	7	NO[c]	Hx	>100	>100	>100

[a] Number of atoms between the N^9-atom of the purine and phosphonate group.
[b] Location of the oxygen from the N^9-atom of the purine ring.
[c] NO = Acyclic linkers without an oxygen atom.
[d] ND = not determined.
[e] G = Guanine, Hx = Hypoxanthine.

of an oxygen atom in the linker always favors *Pf*HGXPRT over the human enzyme. This suggests that these compounds, by contrast with human HGPRT, are better able to induce conformational changes in *Pf*HGXPRT, thereby resulting in enhanced binding affinity.

The optimum length in the number of atoms in the linker for *Pf*HGXPRT appears to be five with guanine as the preferred base. This length is equivalent to the number of atoms in the linker for the naturally occurring products of the reaction, GMP and IMP. Compounds **3g** and **3f** differ from GMP and IMP in two aspects: the absence of the ribose ring, and the substitution of two oxygen atoms for two carbon atoms in the linker. The guanine derivative (**3g**) binds 5–10 times more tightly to the human and *Pf* enzymes than does GMP (5.8 μM for human, 10 μM for *Pf*)[5] while the hypoxanthine derivative (**3f**) binds 6–20 times more weakly to the enzymes than does IMP. Thus, in the case of **3g**, the removal of the ribose ring and substitution of the oxygen atoms for carbon atoms has a net effect of enhancing binding. This may be a result of increased flexibility in the linker due to the removal of the ribose ring. Substitution of the purine base in the PEE compounds does reduce affinity as expected for bases that bind more weakly than the naturally occurring bases, hypoxanthine and guanine.

Compounds with six or seven atoms in the linker region can bind with low K_i values ($\leqslant$0.4 μM) to the human enzyme, but bind much more weakly to the *Pf* enzyme. (>29 μM).This result highlights again that there are clear differences in the ability of the ANPs to bind to the human, *Pf* and *Pv* enzymes. Thus compounds where the linker is $\geqslant$ six atoms will not confer potency or selectivity for the *Plasmodium* enzymes.

ANPs with 3, 4, 5 or atoms in the linker but containing xanthine as the purine base did not inhibit human HGPRT (K_i values >200 μM). The addition of an oxygen in the 2-position did not result in inhibition. This is as expected as xanthine is not a substrate for this enzyme. Thus, the phosphonate moiety alone is not sufficient to anchor the ANP in the active site. These derivatives also did not inhibit the parasite enzymes. Though xanthine is a substrate for *Pf*HGXPRT with a k_{cat} 10 times higher than for hypoxanthine, it binds very weakly (*cf.* K_m for xanthine of 189 μM with 0.07 μM for Hx). When the base is 6-bromopurine, 2-amino-6-bromopurine, 6-chloropurine or 2-amino-6-chloropurine, the compounds also have little or no affinity for either of these two enzymes (K_i values >100 μM). Thus, this data suggests that, for these ANP derivatives to bind tightly, the purine base itself must also be a good substrate.

3. Conclusion

A series of linear alkyl and alkyloxyalkyl phosphonates were synthesised and evaluated as inhibitors of *Pf*HGXPRT and human HGPRT. This study showed that for low K_i values for the ANPs, the purine base must be a good substrate with K_m values <5 μM. Compounds where the linker is too short (1–3 atoms) or too long (>5) do not inhibit the *Plasmodium* enzymes. The position of the oxygen atom in the linker also contributes to their affinity. Optimum binding to the malarial enzymes occurs when the linear linker contains five atoms and the oxygen is located in the 3-position distal from the N^9 atom in the purine ring.

4. Experimental part

4.1. Chemistry

Unless otherwise stated, solvents were evaporated at 40 °C/2 kPa, and the compounds were dried over P_2O_5 at 2 kPa. Solvents were dried by standard procedures. Tetrahydrofuran (THF) was freshly distilled from sodium/benzophenone under argon. Dimethylformamide and acetonitrile were distilled from P_2O_5 and stored over molecular sieves (4 Å). TLC was performed on plates of Kieselgel 60 F254 (Merck). Mass spectra were measured on a LCQ classic spectrometer using electrospray ionization (ESI). 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. The chemicals were obtained from commercial sources (Sigma–Aldrich) or prepared according to the publisher's

procedures. Preparative HPLC purifications were performed on columns packed with 10 μm C18 reversed phase resin (Phenomenex Gemini 10 μm 21 × 250 mm) on Waters Delta 600 chromatograph system; 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 *Pf*HGXPRT, *Pv*HGXPRT and human HGPRT were expressed and purified as previously described.[22] The K_i values were determined by a spectrophotometric method as also described previously.

4.1.1. General procedure: cleavage of the phosphonate diesters with keeping of the 6-chloro group—Method A

The appropriate diester (2 mmol) in CH_3CN (20 mL) and 2,6-lutidine (1.77 mL, 15.2 mmol) were treated dropwise with bromotrimethylsilane (2 mL, 15.2 mmol). The reaction mixture was stirred overnight at room temperature. After evaporation of all volatile materials the residue was codistilled with CH_3CN. The solid was dissolved in H_2O/MeOH 1:1 and adjusted by 1 M HCl to pH 2. The compounds were subsequently purified by HPLC and crystallized.

4.1.2. General procedure: the acid hydrolysis of 6-chloropurine diester—Method B

The appropriate 6-chloropurine diester (3 mmol) was dissolved in 1 M HCl (5 mL) and the reaction mixture was refluxed. The resultant solution was cooled down, neutralized by 1 M NaOH and applied to a Dowex 50 column. Inorganic salts were removed by water and the desired content was eluted with 2.5% aqueous ammonia in 20% solution of MeOH. The compounds were crystallized or purified by column chromatography (SiO_2, 40 g EtOAc/EtOH/Acetone/H_2O 6:1:1:0.5).

4.1.3. General procedure: the basic hydrolysis of 6-chloropurine diester—Method C[18]

The appropriate 6-chloropurine diester (3 mmol) was dissolved or suspended in H_2O (40 mL) containing K_2CO_3 1.69 g (12.2 mmol) and DABCO (1,4-diazabicyclo[2,2,2]octane) 0.69 g (6.2 mmol) and the reaction mixture was refluxed. The resultant solution was cooled down, neutralized by 1 M HCl and extracted to $CHCl_3$. Compounds were purified by column chromatography (SiO_2, 40 g).

4.1.4. General procedure: cleavage of the phosphonate diesters—Method D

The appropriate diester (2 mmol) in CH_3CN (20 mL) DMF (10 mL) was treated dropwise with bromotrimethylsilane (2 mL). The reaction mixture was stirred overnight at room temperature. After evaporation of all volatile materials the residue was codistilled with toluene (3 × 30 mL). The solid was dissolved in 2.5% aqueous NH_3 and applied on the column of Dowex 1 washed with water and eluted with either acetic acid or formic acid. UV absorbing fractions were collected, evaporated and crystallized.

4.1.5. General procedure: diazotation reaction—Method E

Aqueous $NaNO_2$ (12.5 mmol) in 5 mL H_2O was added to the solution of the appropriate guanine derivative 4.2 mmol in HCl 1 M (76 mL). The reaction mixture was stirred for an additional 30 min and neutralized with NaOH. The solvent was evaporated in vacuo and the residue was extracted with 10% MeOH in $CHCl_3$. The filtrate was evaporated and purified by column chromatography (ethyl acetate/ethanol/acetone/H_2O 6:1:1:0.5).

4.1.6. General procedure: diazotation reaction—Method E2

The appropriate adenine compound (1.2 mmol) was dissolved in 80% acetic acid (250 mL) followed by the addition of amyl nitrite (4 mL). The reaction was set aside at room temperature overnight. The mixture was evaporated *in vacuo* and the residue was codistilled with water. The compounds were purified by preparative HPLC chromatography with the product eluted using a linear gradient of H_2O/MeOH (98:20) to (20:80).

4.1.7. General procedure: alkylation of 2-amino-6-chloropurine—Method F

The mixture of the appropriate halopurine (23 mmol) in DMF (50 mL) was treated with NaH (60% in mineral oil) (25.8 mmol) for 1 h at room temperature. Alkyl phosphonate ester (25.8 mmol) in DMF (20 mL) was added to the solution and the resultant mixture was stirred at 80 °C. The product was evaporated in vacuo and codistilled with toluene three times. The residue was extracted by $CHCl_3$. The resulting extract was concentrated, applied on the column of silicagel and eluted by MeOH/$CHCl_3$ gradient.

4.1.8. General procedure: alkylation of 2-amino-6-chloropurine—Method G

The mixture of the appropriate halopurine (8.5 mmol) in DMF (30 mL) was treated with NaH (60% in mineral oil, 10.6 mmol) for 1 h at room temperature. The reaction mixture was cooled down to −10 °C and the appropriate alkyl phosphonate ester (8.5 mmol) in DMF (10 mL) was added to the solution. The resultant mixture was allowed to warm to room temperature and stirred at this temperature for 24–72 h. The product was evaporated in vacuo and codistilled three times with toluene. The residue was extracted with $CHCl_3$. The resulting extract was concentrated, applied on the column of silica gel and eluted by MeOH/$CHCl_3$ gradient.

4.1.9. General procedure: cleavage of the phosphonate diesters—Method H

The appropriate diester (2 mmol) in CH_3CN (20 mL) was treated dropwise with bromotrimethylsilane (2 mL). The reaction mixture was stirred overnight at room temperature. After evaporation of all volatile materials the residue was codistilled with CH_3CN. The solid was dissolved in H_2O and neutralized by Dowex 50 in Na^+ cycle. Compounds were subsequently purified by crystallization or by HPLC.

4.1.9.1. Diisopropyl 2-amino-6-chloro-9-(2-phosphonomethyl)-9*H*-purine (1a). Method F: 2-amino-6-chloropurine 2 g (11.8 mmol); diisopropyl bromomethylphosphonate (**8a**) 3 mL (12 mmol); 80 °C 8 h; column chromatography 5–10% MeOH in $CHCl_3$; crystallized EtOH; obtained 1.8 g (44%) of compound **1a**. ESI [M+H] 348.1 (100).^{1}H NMR ($CDCl_3$): 7.99 (s, 1H, H-8); 5.18 (bs, 2H, NH_2); 4.72 (dm, 2H, $J_{H-C-O-P}$ = 7.2 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 4.41 (d, 2H, J_{H-C-P} = 12.3 Hz, H-1′); 1.29 and 1.22 (2 × d, 2 × 6H, J_{CH3-CH} = 6.2 Hz, CH_3-*i*Pr); ^{13}C NMR ($CDCl_3$): 158.97 (C-2); 153.67 (d, J_{C4-P} = 3.1 Hz, C-4); 151.19 (C-6); 142.42 (C-8); 124.37 (C-5); 72.54 (d, J_{C-O-P} = 6.9 Hz, CH-*i*Pr); 39.49 (d, J_{C-P} = 158.9 Hz, C-1′); 23.95 and 23.82 (2 × d, $J_{C-C-O-P}$ = 3.9 Hz, $J_{C-C-O-P}$ = 4.8 Hz, CH_3-*i*Pr). For $C_{12}H_{19}ClN_5O_3P$ (347.74) calcd: C, 41.45; H, 5.51; N, 20.14; P, 8.91. Found: C, 41.31; H, 5.45; N, 20.05; P, 8.74.

4.1.9.2. Diisopropyl 6-chloro-9-(3-phosphonopropyl)-9*H*-purine (1b). Method F: 6-chloropurine, 3 g (19 mmol); diisopropyl 3-bromopropylphosphonate (**8b**); 80 °C, 5 h; column chromatography

0–1% MeOH in $CHCl_3$; obtained 4.96 g (71%) of compound **1b**. ESI [M+Na] 383.1 (100).^{1}H NMR (DMSO-d_6): 8.78 (s, 1H, H-2); 8.71 (s, 1H, H-8); 4.51 (dn, 2H, $J_{H–C–O–P}$ = 8.0 Hz, $J_{CH–CH3}$ = 6.2 Hz, CH-*i*Pr); 4.35 (t, 2H, $J_{1′-2′}$ = 7.1 Hz, H-1′); 2.05 (m, 2H, H-2′); 1.70 (dm, 2H, $J_{3′-P}$ = 18.5 Hz, H-3′); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.18 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 152.22 (C-4); 151.67 (C-2); 149.21 (C-6); 147.79 (C-8); 131.18 (C-5); 69.60 (d, $J_{C–O–P}$ = 6.4 Hz, CH-*i*Pr); 44.38 (d, $J_{1′-P}$ = 19.3 Hz, C-1′); 23.95 (m, CH_3-*i*Pr); 23.33 (d, $J_{3′-P}$ = 141.8 Hz, C-3′); 23.01 (d, $J_{2′-P}$ = 4.5 Hz, C-2′).

4.1.9.3. Diisopropyl 2-amino-6-chloro-9-(3-phosphonopropyl)-9*H*-purine (1c). Method F: 2-amino-6-chloropurine 5 g (29 mmol); diisopropyl 3-bromopropylphosphonate (**8b**); 80 °C 8 h; column chromatography 0–2% MeOH in $CHCl_3$; crystallized $CHCl_3$ acetone; obtained 6.81 g (61%) of compound **1c**. ESI [M+Na] 398.0 (60).^{1}H NMR (DMSO-d_6): 8.12 (s, 1H, H-8); 6.92 (br s, 2H, NH_2); 4.51 (dn, 2H, $J_{H–C–O–P}$ = 8.0 Hz, $J_{CH–CH3}$ = 6.2 Hz, CH-*i*Pr); 4.09 (t, 2H, $J_{1′-2′}$ = 7.0 Hz, H-1′); 1.95 (m, 2H, H-2′); 1.63 (dm, 2H, $J_{3′-P}$ = 18.4 Hz, H-3′); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.18 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 159.99 (C-2); 154.28 (C-4); 149.60 (C-6); 143.53 (C-8); 123.63 (C-5); 69.62 (d, $J_{C–O–P}$ = 6.4 Hz, CH-*i*Pr); 43.59 (d, $J_{1′-P}$ = 19.8 Hz, C-1′); 23.95 (m, CH_3-*i*Pr); 23.39 (d, $J_{3′-P}$ = 142.0 Hz, C-3′); 22.82 (d, $J_{2′-P}$ = 4.4 Hz, C-2′). For $C_{14}H_{23}ClN_5O_3P$ (375.79) calcd: C, 44.75; H, 6.17; N, 18.64; P, 8.24. Found: C, 44.76; H, 5.99; N, 18.31; P, 8.57.

4.1.9.4. Diethyl 6-chloro-9-(4-phosphonobutyl)-9*H*-purine (1d). Method F: 6-chloropurine 3 g (19.4 mmol); diisopropyl 4-bromobutylphosphonate[10,12] (**8c**); 80 °C; 2.5 h; column chromatography 1–2% MeOH in $CHCl_3$ crystallized from $CHCl_3$ acetone; obtained 4.2 g (62%) of compound **1d**. ESI [M+H] 347.0 (100).^{1}H NMR (DMSO-d_6): 8.78 (s, 1H, H-2); 8.73 (s, 1H, H-8); 4.32 (t, 2H, $J_{1′-2′}$ = 6.9 Hz, H-1′); 3.88–3.96 (m, 4H, CH_2–O); 1.95 (m, 2H, H-2′); 1.76 (dm, 2H, $J_{C–H–P}$ = 15.3 Hz H-4′); 1.41 (m, 2H, H-3′); 1.16 (t, 6H, $J_{CH3,H2}$ = 7.1 Hz, CH_3). ^{13}C NMR (DMSO-d_6): 152.18 (C-4); 151.69 (C-2); 149.21 (C-6); 147.78 (C-8); 131.03 (C-5); 61.04 (d, $J_{C–O–P}$= 6.3 Hz CH_2–O); 43.44 (C-1′); 29.79 (d, $J_{2′-P}$ = 16.2 Hz, C-2′); 23.93 (d, $J_{4′-P}$ = 138.8 Hz, C-4′); 19.41 (d, $J_{3′-P}$ = 4.9 Hz, C-3′); 16.45 (d, $J_{C–C–O–P}$ = 5.7 Hz CH_3).

4.1.9.5. Diisopropyl 2-amino-6-chloro-9-(4-phosphonobutyl)-9*H*-purine (1e). Method F: 2-amino-6-chloropurine 4 g (23 mmol); diisopropyl 4-bromobutylphosphonate[10,12] (**8c**); 0 °C; 5 h; column chromatography 0–4% MeOH in $CHCl_3$; crystallized from $CHCl_3$ acetone; obtained 5.9 g (64%) of compound **1e**. ESI [M+H] 390.2 (100).^{1}H NMR (DMSO-d_6): 8.13 (s, 1H, H-8); 6.89 (br s, 2H, NH_2); 4.49 (dn, $J_{C–C–O–P}$ = 8.0 Hz, $J_{CH3,CH}$ = 6.2 Hz, CH-*i*Pr); 4.06 (t, 2H, $J_{1′-2′}$ = 6.9 Hz, H-1′); 1.85 (m, 2H, H-2′); 1.68 (dm, $J_{4′-P}$ = 18.1 Hz, 2H, H-4′); 1.38 (m, 2H, H-3′); 1.19 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.16 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6):159.95 (C-2); 154.28 (C-4); 149.52 (C-6); 143.44 (C-8); 123.53 (C-5); 69.28 (d, $J_{C–O–P}$ = 6.4 Hz, CH-*i*Pr);42.57 (C-1′); 29.69 (d, $J_{2′-P}$ = 16.3 Hz, C-2′); 25.33 (d, $J_{4′-P}$ = 140.5 Hz, C-4′); 19.54 (d, $J_{3′-P}$ = 5.0 Hz, C-3′); 23.93 (d, $J_{C–C–O–P}$ = 4.0 Hz, CH_3-*i*Pr); 23.89 (d, $J_{C–C–O–P}$ = 4.8 Hz, CH_3-*i*Pr). For $C_9H_{13}BrN_5O_4P$ (389.82) calcd: C, 46.22; H, 6.46; Cl, 9.09; N, 17.97; P, 7.95. Found: C, 46.07; H, 6.53; Cl, 9.18; N, 17.62; P, 8.15.

4.1.9.6. Diisopropyl 6-chloro-9-(4-phosphonopentyl)-9*H*-purine (1f).Method F: 6-chloropurine 3 g (19.4 mmol); diisopropyl 5-bromopentylphosphonate (**8d**) prepared according lit. (MW, 40 W, 130 °C, 1 h)[12]) 80 °C, 5 h, column chromatography 0–2% MeOH in $CHCl_3$; obtained 5.57 g (74%) of compound **1f**. ESI [M+Na] 411.8 (100).^{1}H NMR (DMSO-d_6): 8.77 (s, 1H, H-2); 8.73 (s, 1H, H-8); 4.49 (dn, 2H, $J_{H–C–O–P}$ = 8.0 Hz, $J_{CH–CH3}$ = 6.2 Hz, CH-*i*Pr); 4.28 (t, 2H, $J_{1′-2′}$ = 7.1 Hz, H-1′); 1.87 (m, 2H, H-2′); 1.61 (m, 2H, H-5′); 1.45 (m, 2H, H-4′); 1.30 (m, 2H, H-3′); 1.19 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.18 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 152.21 (C-4); 151.66 (C-2); 149.17 (C-6); 147.77 (C-8); 131.03 (C-5); 69.20 (d, $J_{C–O–P}$ = 6.3 Hz, CH-*i*Pr); 43.86 (C-1′); 28.77 (C-2′); 26.89 (d, $J_{3′-P}$ = 16.0 Hz, C-3′); 25.83 (d, $J_{5′-P}$ = 140.1 Hz, C-5′); 23.97 (m, CH_3-*i*Pr); 22.06 (d, $J_{4′-P}$ = 5.1 Hz, C-4′). For $C_{16}H_{26}ClN_4O_3P$ (388.83) calcd: C, 49.42; H, 6.74; N, 14.41; Cl, 9.12. Found: C, 49.15; H, 7.10; N, 14.05; Cl, 9.30.

4.1.9.7. Diisopropyl 2-amino-6-chloro-9-(4-phosphonopentyl)-9*H*-purine (1g). Method F: 2-amino-6-chloropurine 4 g (23.5 mmol); diisopropyl 5-bromopentylphosphonate (**8d**); 80 °C, 4 h, column chromatography 0–1% MeOH in $CHCl_3$; obtained 7.25 g (76%) of compound **1g**. ESI [M+Na] 426.8 (100).^{1}H NMR (DMSO-d_6): 8.14 (s, 1H, H-8); 6.91 (br s, 2H, NH_2); 4.50 (dn, 2H, $J_{H–C–O–P}$ = 8.0 Hz, $J_{CH–CH3}$ = 6.2 Hz, CH-*i*Pr); 4.02 (t, 2H, $J_{1′-2′}$ = 7.1 Hz, H-1′); 1.77 (m, 2H, H-2′); 1.62 (m, 2H, H-5′); 1.45 (m, 2H, H-4′); 1.29 (m, 2H, H-3′); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.1 Hz, CH_3-*i*Pr); 1.19 (d, 6H, $J_{CH3,CH}$ = 6.1 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 159.96 (C-2); 154.31 (C-4); 149.50 (C-6); 143.47 (C-8); 123.55 (C-5); 69.21 (d, $J_{C–O–P}$ = 6.5 Hz, CH-*i*Pr); 43.06 (C-1′); 28.74 (C-2′); 27.01 (d, $J_{3′-P}$ = 16.1 Hz, C-3′); 25.90 (d,$J_{5′-P}$ = 140.0 Hz, C-5′); 23.98 (m, $J_{C–C–O–P}$– 4.0 Hz, CH_3-*i*Pr); 22.11 (d, $J_{4′-P}$ = 5.1 Hz, C-4′). For $C_{16}H_{27}ClN_5O_3P{\cdot}2/3\ H_2O$ (403.84) calcd: C, 46.21; H, 6.87; N, 16.84. Found: C, 46.50; H, 6.82; N, 16.60.

4.1.9.8. Diisopropyl 6-chloro-9-(6-phosphonohexyl)-9*H*-purine (1h). Method F: 6-chloropurine 2 g (13 mmol); diisopropyl 6-chlorohexylphosphonate (**8e**); 80 °C, 30 h; column chromatography 0–2% MeOH in $CHCl_3$; obtained 1.39 g (27%) of compound **1h**. ESI [M+H] 403.1 (100).^{1}H NMR (DMSO-d_6): 8.77 (s, 1H, H-2); 8.72 (s, 1H, H-8); 4.51 (dn, 2H, $J_{H–C–O–P}$ = 8.0 Hz, $J_{CH–CH3}$ = 6.2 Hz, CH-*i*Pr); 4.28 (t, 2H, $J_{1′-2′}$ = 7.1 Hz, H-1′); 1.84 (m, 2H, H-2′); 1.59 (m, 2H, H-6′); 1.35–1.45 (m, 4H, H-4′, 5′); 1.24 (m, 2H, H-3′); 1.21 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 152.17 (C-4); 151.62 (C-2); 149.16 (C-6); 147.77 (C-8); 131.04 (C-5); 69.17 (d, $J_{C–O–P}$ = 6.4 Hz, CH-*i*Pr); 43.97 (C-1′); 29.28 (d, $J_{4′-P}$ = 16.3 Hz, C-4′); 28.99 (C-2′); 25.88 (d, $J_{6′-P}$ = 140.1 Hz, C-6′); 25.63 (C-3′); 23.98 (m, CH_3-*i*Pr); 22.25 (d, $J_{5′-P}$ = 5.2 Hz, C-5′).

4.1.9.9. Diisopropyl 2-amino-6-chloro-9-(6-phosphonohexyl)-9*H*-purine (1i).Method F: 2-amino-6-chloropurine 4 g (23.5 mmol); diisopropyl 6-chlorohexylphosphonate (**8e**); 110 °C 24 h; column chromatography 0–2% MeOH in $CHCl_3$; obtained 3.6 g (37%) of compound **1i**. ESI [M+H] 415.1 (100).^{1}H NMR (DMSO-d_6): 8.13 (s, 1H, H-2); 6.89 (br s, 2H, NH_2); 4.51 (dn, 2H, $J_{H–C–O–P}$ = 8.0 Hz, $J_{CH–CH3}$ = 6.2 Hz, CH-*i*Pr); 4.02 (t, 2H, $J_{1′-2′}$ = 7.1 Hz, H-1′); 1.75 (m, 2H, H-2′); 1.61 (m, 2H, H-6′); 1.32–1.46 (m, 4H, H-4′, 5′); 1.23 (m, 2H, H-3′); 1.21 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 159.95 (C-2); 154.28 (C-4); 149.49 (C-6); 143.50 (C-8); 123.54 (C-5); 69.20 (d, $J_{C–O–P}$ = 6.2 Hz, CH-*i*Pr); 43.09 (C-1′); 29.31 (d, $J_{4′-P}$ = 6.2 Hz, C-4′); 28.93 (C-2′); 25.89 (d, $J_{6′-P}$ = 140.0 Hz, C-6′); 25.66 (C-3′); 23.99 (m, CH_3-*i*Pr); 22.29 (d, $J_{5′-P}$ = 5.2 Hz, C-5′).

4.1.9.10. Diisopropyl 6-chloro-9-[7-(phosphonoheptyl]-9*H*-purine (1j). Method F: 6-chloropurine 1.5 g (9.7 mmol); diisopropyl 7-bromoheptylphosphonate (**8f**); 6 h 80 °C; column chromatography 0–2% MeOH in $CHCl_3$; obtained 2.3 g (57%) of compound **1j**. ESI [M+H] 417.1 (80).^{1}H NMR (DMSO-d_6): 8.77 (s, 1H, H-2); 8.73 (s, 1H, H-8); 4.51 (m, 2H, CH-*i*Pr); 4.29 (t, 2H, $J_{1′-2′}$ = 7.1 Hz, H-1′); 1.85 (m, 2H, H-2′); 1.61 (m, 2H, H-7′); 1.41 (m, 2H, H-6′); 1.24-1.33 (m, 4H, H-4′, 5′); 1.20 (m, 2H, H-3′); 1.20 (d, 6H, $J_{CH3,\ CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.19 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr).

^{13}C NMR (DMSO-d_6):152.20 (C-4); 151.69 (C-2); 149.20 (C-6); 147.81 (C-8); 131.04 (C-5); 69.18 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 43.98 (C-1′); 29.76 (d, $J_{5'-P}$ = 16.3 Hz, C-5′); 29.18 (C-2′); 28.06 (C-4′); 25.96 (d, $J_{7'-P}$ = 139.9 Hz, C-7′); 25.93 (C-3′); 24.03 (m, CH_3-*i*Pr); 22.30 (d, $J_{6'-P}$ = 5.2 Hz, C-6′).

4.1.9.11. Diisopropyl 2-amino-6-chloro-9-[7-(phosphonoheptyl] -9*H*-purine (1k). Method F: 2-amino-6-chloropurine 3 g (18 mmol); diisopropyl 7-bromoheptylphosphonate (**8f**); 7 h 80 °C; column chromatography 0–3% MeOH in $CHCl_3$; obtained 3.39 g (44%) of compound **1k**. ESI [M+H] 432.2 (85).^{1}H NMR (DMSO-d_6): 8.14 (s, 1H, H-8); 6.91 (br s, 2H, NH_2); 4.51 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 4.03 (t, 2H, $J_{1'-2'}$ = 7.1 Hz, H-1′); 1.75 (m, 2H, H-2′); 1.61 (m, 2H, H-7′); 1.41 (m, 2H, H-6′); 1.24-1.33 (m, 4H, H-4′, 5′); 1.20 (m, 2H, H-3′); 1.21 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 159.98 (C-2); 154.30 (C-4); 149.53 (C-6); 143.55 (C-8); 123.55 (C-5); 69.20 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 43.20 (C-1′); 29.81 (d, $J_{5'-P}$ = 16.1 Hz, C-5′); 29.09 (C-2′); 28.17 (C-4′); 26.01 (C-3′); 25.98 (d, $J_{7'-P}$ = 139.9 Hz, C-7′); 24.03 (m, CH_3-*i*Pr); 22.35 (d, $J_{6'-P}$ = 5.1 Hz, C-6′).

4.1.9.12. Diisopropyl 9-(3-phosphonopropyl)hypoxanthine (2b). Method B: compound **1b**, 1.5 g (9.1 mmol); 6 h reflux; obtained 1.35 g (95%) of compound **2b**. ESI [M−H] 342.3 (100).^{1}H NMR (DMSO-d_6): 8.08 (s, 1H, H-8); 8.03 (s, 1H, H-2); 4.51 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 4.19 (t, 2H, $J_{1'-2'}$ = 7.0 Hz, H-1′); 1.96 (m, 2H, H-2′); 1.63 (m, 2H, H-3′); 1.21 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.19 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 156.90 (C-6); 148.55 (C-4); 145.73 (C-2); 140.56 (C-8); 124.26 (C-5); 69.61 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 43.82 (d, $J_{1'-P}$ = 19.5 Hz, C-1′); 23.93 (m, CH_3-*i*Pr); 23.53 (d, $J_{2'-P}$ = 4.5 Hz, C-2′); 23.35 (d, $J_{3'-P}$ = 141.4 Hz, C-3′).

4.1.9.13. Diisopropyl 9-(3-phosphonopropyl)guanine (2c). Method B: Compound **1c**, 4.48 g (12 mmol); reflux 8 h, crystallized H_2O; obtained 3.96 g (93%) of compound **2c**. ESI [M+Na] 380.1 (100).^{1}H NMR (DMSO-d_6): 10.59 (br s, 1H, NH); 7.68 (s, 1H, H-8); 6.46 (br s, 2H, NH_2); 4.51 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.98 (t, 2H, $J_{1'-2'}$ = 6.9 Hz, H-1′); 1.89 (m, 2H, H-2′); 1.59 (dm, 2H, $J_{3'-P}$ = 18.2 Hz, H-3′); 1.21 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.19 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 157.08 (C-6); 153.78 (C-2); 151.36 (C-4); 137.78 (C-8); 116.89 (C-5); 69.64 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 43.21 (d, $J_{1'-P}$ = 20.0 Hz, C-1′); 23.96 (m, CH_3-*i*Pr); 23.38 (d, $J_{3'-P}$ = 142.0 Hz, C-3′); 23.28 (d, $J_{2'-P}$ = 4.3 Hz, C-2′). For $C_{14}H_{24}N_5O_4P$ (357.35). 4/3 H_2O calcd: C, 44.09; H, 7.05; N, 18.36; P, 8.12. Found: C, 44.13; H, 6.97; N, 18.25; P, 8.43.

4.1.9.14. Diethyl 9-(4-phosphonobutyl)hypoxanthine (2d). Method C: compound **1d**, 3.11 g (9 mmol); Reflux 2.5 h, neutralized Dowex 50, extracted 5% MeOH in $CHCl_3$; compound **1d** was partially deprotected to the phosphonate monoester under these conditions and used without another purification and characterization in the next step.

4.1.9.15. Diisopropyl 9-(4-phosphonobutyl)guanine (2e). Method B: compound **1e**, 5.0 g (12.8 mmol); reflux 4 h; column chromatography; crystallized EtOH; obtained 3.65 g (77%) of compound **2e**. ESI [M+H] 372.1 (100). ^{1}H NMR (DMSO-d_6): 10.53 (br s, 1H, NH); 7.67 (s, 1H, H-8); 6.41 (br s, 2H, NH_2); 4.50 (dn, $J_{C-C-O-P}$ = 8.0 Hz, $J_{CH3,CH}$ = 6.2 Hz, CH-*i*Pr); 3.94 (t, 2H, $J_{1'-2'}$ = 6.9 Hz, H-1′); 1.79 (m, 2H, H-2′); 1.68 (dm, 2H, $J_{4'-P}$ = 18.0 Hz, H-4′); 1.38 (m, 2H, H-3′); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.18 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 157.05 (C-6); 153.68 (C-2); 151.37 (C-4); 137.65 (C-8); 116.76 (C-5); 69.29 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 42.19 (C-1′); 30.19 (d, $J_{2'-P}$ = 16.2 Hz, C-2′); 25.38 (d, $J_{4'-P}$ = 140.4 Hz, C-4′); 23.95 (m, CH_3-*i*Pr); 19.52 (m, $J_{3'-P}$ = 4.9 Hz, C-3′). For $C_{15}H_{26}N_5O_4P$. 1/3 H_2O (371.37) calcd: C, 47.74; H, 7.12; N, 18.56; P, 8.21. Found: C, 47.74; H, 6.99; N, 18.37; P, 8.56.

4.1.9.16. Diisopropyl 9-(5-phosphonopentyl)hypoxanthine (2f). Method B: Compound **1f**, 3.57 g (9.1 mmol); reflux 4 h; obtained 2 g (59%) of compound **2f**. ESI [M+Na] 393.2 (100).^{1}H NMR (DMSO-d_6): 12.26 (br s, 1H, NH); 8.09 (s, 1H, H-8); 8.02 (s, 1H, H-2); 4.50 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 4.11 (t, 2H, $J_{1'-2'}$ = 7.1 Hz, H-1′); 1.79 (m, 2H, H-2′); 1.62 (m, 2H, H-5′); 1.45 (m, 2H, H-4″.29 (m, 2H, H-3′); 1.19 (m, 12H, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 156.93 (C-6); 148.60 (C-4); 145.62 (C-2); 140.51 (C-8); 124.13 (C-5); 69.22 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 43.32 (C-1′); 29.32 (C-2′); 26.93 (d, $J_{3'-P}$ = 16.2 Hz, C-3′); 25.88 (d, $J_{5'-P}$ = 140.0 Hz, C-5′); 24.00 (d, $J_{C-C-O-P}$ = 4.0 Hz, CH_3-*i*Pr); 22.07 (d, $J_{4'-P}$ = 4.9 Hz, C-4′). For $C_{16}H_{27}N_4O_4P$ (370.38) calcd: C, 51.89; H, 7.39; N, 15.13; P, 8.36. Found: C, 51.78; H, 7.39; N, 15.03; P, 8.70.

4.1.9.17. Diisopropyl 9-(5-phosphonopentyl)guanine (2 g). Method B: Compound **1g**, 5.8 g (14.3 mmol); reflux 4 h; obtained 4.0 g (65%) of compound **2g**. ESI [M+Na] 408.39 (50).^{1}H NMR (DMSO-d_6): 7.63 (s, 1H, H-8); 6.72 (br s, 2H, NH_2); 4.51 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.89 (t, 2H, $J_{1'-2'}$ = 7.1 Hz, H-1′); 1.71 (m, 2H, H-2′); 1.63 (m, 2H, H-5′); 1.46 (m, 2H, H-4′); 1.29 (m, 2H, H-3′); 1.21 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 158.81 (C-6); 155.00 (C-2); 151.57 (C-4); 137.24 (C-8); 116.83 (C-5); 69.28 (d, J_{C-O-P} = 6.3 Hz, CH-*i*Pr); 42.61 (C-1′); 29.29 (C-2′); 27.09 (d, $J_{3'-P}$ = 16.3 Hz, C-3′); 25.94 (d, $J_{5'-P}$ = 140.0 Hz, C-5′); 24.03 (d, $J_{C-C-O-P}$– 3.5 Hz, CH_3-*i*Pr); 22.16 (d, $J_{4'-P}$ = 4.7 Hz, C-4′).

4.1.9.18. Diisopropyl 9-(6-phosphonohexyl]hypoxanthine (2 h). Method B: Compound **1h**, 0.86 g (2.1 mmol); reflux 6 h; obtained 0.44 g (54%) of compound **2h**. ESI [M+Na] 407.2 (100).^{1}H NMR (DMSO-d_6): 8.08 (s, 1H, H-8); 8.02 (s, 1H, H-2); 4.51 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 4.12 (t, 2H, $J_{1'-2'}$ = 7.1 Hz, H-1′); 1.76 (m, 2H, H-2′); 1.60 (m, 2H, H-6′); 1.40 (m, 2H, H-5′); 1.35 (m, 2H, H-4′); 1.22 (m, 2H, H-3′); 1.21 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 156.92 (C-6); 148.57 (C-4); 145.60 (C-2); 140.53 (C-8); 124.14 (C-5); 69.21 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 43.42 (C-1′); 29.53 (C-2′); 29.29 (d, $J_{4'-P}$ = 16.3 Hz, C-4′); 25.89 (d, $J_{6'-P}$ = 140.1 Hz, C-6′); 25.62 (C-3′); 24.00 (d, $J_{C-C-O-P}$ = 4.1 Hz, CH_3-*i*Pr); 22.29 (d, $J_{5'-P}$ = 5.2 Hz, C-5′).

4.1.9.19. Diisopropyl 9-(6-phosphonohexyl)guanine (2i). Method B: Compound **1i**, 3.1 g (7.4 mmol); reflux 6 h; column chromatography (MeOH:$CHCl_3$ 5:95); obtained 1.43 g (48%) of compound **2i**. ESI [M+Na] 422.2 (100).^{1}H NMR (DMSO-d_6): 10.52 (br s, 1H, NH); 7.67 (s, 1H, H-8); 6.42 (br s, 2H, NH_2); 4.52 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.91 (t, 2H, $J_{1'-2'}$ = 7.1 Hz, H-1′); 1.69 (m, 2H, H-2′); 1.61 (m, 2H, H-6′); 1.43 (m, 2H, H-5′); 1.35 (m, 2H, H-4′); 1.22 (m, 2H, H-3′); 1.22 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.21 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 157.04 (C-6); 153.66 (C-2); 151.35 (C-4); 137.68 (C-8); 116.76 (C-5); 69.21 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 42.71 (C-1′); 29.41 (C-2′); 29.32 (d, $J_{4'-P}$ = 16.3 Hz, C-4′); 25.90 (d, $J_{6'-P}$ = 140.1 Hz, C-6′); 25.65 (C-3′); 24.00 (d, $J_{C-C-O-P}$ = 4.1 Hz, CH_3-*i*Pr); 22.31 (d, $J_{5'-P}$ = 5.2 Hz, C-5′).

4.1.9.20. Diisopropyl 9-(7-phosphonoheptyl)hypoxanthine (2j). Method C: Compound **1j**, 1.06 g (2.5 mmol); reflux 2 h; obtained 1.0 g (98%) of compound **2j**. ESI [M+Na] 421.3 (100).^{1}H NMR (DMSO-d_6): 8.09 (s, 1H, H-8); 8.02 (s, 1H, H-2); 4.51 (m, 2H, CH-*i*Pr); 4.12 (t, 2H, $J_{1'-2'}$ = 7.1 Hz, H-1′); 1.77 (m, 2H, H-2′);

1.60 (m, 2H, H-7′); 1.41 (m, 2H, H-6′); 1.24-1.34 (m, 4H, H-4′, 5′); 1.20 (m, 2H, H-3′); 1.21 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 156.93 (C-6); 148.61 (C-4); 145.63 (C-2); 140.57 (C-8); 124.13 (C-5); 69.20 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 43.44 (C-1′); 29.82 (d, $J_{5'-P}$ = 16.4 Hz, C-5′); 29.73 (C-2′); 28.11 (C-4′); 25.98 (d, $J_{7'-P}$ = 139.8 Hz, C-7′); 25.94 (C-3′); 24.05 (m, CH_3-*i*Pr); 22.34 (d, $J_{6'-P}$ = 5.2 Hz, C-6′).

4.1.9.21. Diisopropyl 9-[7-(phosphonoheptyl]guanine (2 k). Method B: Compound **1k**, 2.36 g (5.5 mmol); reflux 4 h; column chromatography; obtained 1.58 g (70%) of compound **2k**. ESI [M+Na] 436.3 (100).^{1}H NMR (DMSO-d_6): 10.55 (br s, 1H, NH); 7.67 (s, 1H, H-8); 6.44 (br s, 2H, NH_2); 4.51 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.91 (t, 2H, $J_{1'-2'}$ = 7.2 Hz, H-1′); 1.69 (m, 2H, H-2′); 1.61 (m, 2H, H-7′); 1.41 (m, 2H, H-6′); 1.22-1.33 (m, 4H, H-4′, 5′); 1.20 (m, 2H, H-3′); 1.21 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 157.10 (C-6); 153.69 (C-2); 151.37 (C-4); 137.73 (C-8); 116.78 (C-5); 69.21 (d, J_{C-O-P} = 6.5 Hz, CH-*i*Pr); 42.81 (C-1′); 29.87 (d, $J_{5'-P}$ = 16.1 Hz, C-5′); 29.60 (C-2′); 28.22 (C-4′); 26.02 (C-3′); 25.99 (d, $J_{7'-P}$ = 139.9 Hz, C-7′); 24.04 (m, CH_3-*i*Pr); 22.37 (d, $J_{6'-P}$ = 5.3 Hz, C-6′).

4.1.9.22. 9-(1-phosphonomethyl)guanine (3a). From compound **1a**, 1.8 g (5.18 mmol) by Method C: without purification used for the next step Method D: crystallized H_2O/MeOH; obtained 570 mg (45%) of compound **3a**. ESI [M−H] 244.0 (100). ^{1}H NMR (D_2O): 7.92 (s, 1H, H-8); 3.98 (d, 2H, $J_{(H-C-P)}$ = 12.1 Hz, H-1′); ^{13}C NMR (D_2O): 168.92 (C-6); 161.6 (C-2); 152.37 (d, $J_{(C4-P)}$ = 3.8 Hz), C-4); 139.33 (C-8); 117.50 (C-5); 42.41 (d, $J_{(C-P)}$ = 134.3 Hz, C-1′); For $C_6H_8N_5O_4P$ (245.13) calcd: C, 29.40; H, 3.29; N, 28.57; P, 12.64. Found: C, 29.26; H, 3.60; N, 28.47; P, 12.72.

4.1.9.23. 9-(3-Phosphonopropyl)hypoxanthine (3b). Method D: Compound **2b**, 1.3 g (3.8 mmol); crystallized from H_2O; obtained 0.71 g (72%) of compound **3b**. ESI [M−H] 257.0 (100).^{1}H NMR (D_2O+NaOD): 8.16 (s, 1H, H-8); 8.15 (s, 1H, H-2); 4.27 (t, 2H, $J_{1'-2'}$ = 7.1 Hz, H-1′); 2.07 (m, 2H, H-2′); 1.46 (m, 2H, H-3′). ^{13}C NMR (D_2O+NaOD): 159.30 (C-6); 149.37 (C-4); 146.13 (C-2); 142.91 (C-8); 123.88 (C-5); 45.93 (d, $J_{1'-P}$ = 19.5 Hz, C-1′); 26.20 (d, $J_{3'-P}$ = 132.9 Hz, C-3′); 25.33 (C-2′). For $C_8H_{11}N_4O_4P$. 4/3 H_2O (258.17) calcd: C, 34.05; H, 4.88; N, 19.85; P, 10.98. Found: C, 33.82; H, 4.72; N, 19.64; P, 11.30.

4.1.9.24. 9-(3-phosphonopropyl)guanine (3c). Method D: Compound **2c**, 1.92 g, (5.3 mmol), Dowex 1; 0.5-1 M AcOH and 1 M HCOOH; crystallized from H_2O; obtained 1 g (68%) of compound **3c**. ESI [M−H] 272.0 (100).^{1}H NMR (D_2O+NaOD): 7.81 (s, 1H, H-8); 4.04 (t, 2H, $J_{1'-2'}$ = 7.3 Hz, H-1′); 1.98 (m, 2H, H-2′); 1.38 (m, 2H, H-3′). ^{13}C NMR (DMSO-d_6): 160.48 (C-6); 154.97 (C-2); 151.88 (C-4); 140.63 (C-8); 116.64 (C-5); 45.57 (d, $J_{1'-P}$ = 19.5 Hz, C-1′); 26.85 (d, $J_{3'-P}$ = 130.6 Hz, C-3′); 25.56 (d, $J_{2'-P}$ = 3.2 Hz, C-2′). For $C_8H_{12}N_5O_4P$ (273.19). 2/5 H_2O calcd: C, 34.27; H, 4.60; N, 24.58; P, 11.05. Found: C, 34.54; H, 4.47; N, 24.27; P, 11.06.

4.1.9.25. 9-(4-Phosphonobutyl)hypoxanthine (3d). Method D: Compound **2d**, 0.72 g (2.1 mmol); additional purification on HPLC; crystallized from H_2O; obtained 0.35 g (62%) of compound **3d**. ESI [M−H] = 271.4 (100). ^{1}H NMR (D_2O): 8.65 (s, 1H, H-8); 8.26 (s, 1H, H-2); 4.36 (t, 2H, $J_{1'-2'}$ = 7.0 Hz, H-1′); 2.01 (t, 2H, $J_{2'-1'}$ = $J_{2'-3'}$ = 7.4 Hz, H-2′); 1.74 (m, 2H, H-4′); 1.58 (m, 2H, H-3′); ^{13}C NMR (D_2O): 157.28 (C-6); 148.69 (C-4); 147.71 (C-2); 141.60 (C-8); 120.41 (C-5); 45.38 (C-1′); 30.53 (d, $J_{2'-P}$ = 16.8 Hz, C-2′); 26.89 (d, $J_{4'-P}$ = 134.7 Hz, C-4′); 20.24 (d, $J_{3'-P}$ = 4.5 Hz, C-3′). For $C_9H_{13}N_4O_4P$. 2/3 H_2O (272.20) calcd: C, 38.03; H, 5.08; N, 19.71; P, 10.90. Found: C, 38.05; H, 5.03 N, 19.73; P, 10.90.

4.1.9.26. 9-(4-phosphonobutyl)guanine (3e). Method D: 1.32 g (3.5 mmol) **2e** was eluted from Dowex 50 evaporated, applied on D50 in Na^+ crystallized aqueous EtOH, obtained 0.94 g (81%) of sodium salt **3e**.

Compound **1e**, 0.75 g (1.9 mmol) was hydrolyzed in an attempt to obtain appropriate free halogenopurine phosphonate. The reaction mixture was quenched with H_2O after the codistillation and crystallized from H_2O; obtained 0.35 g (63%) of compound **3e**; The spectrum was in accordance with above mentioned **3e** and lit.[19]

4.1.9.27. 9-(5-Phosphonopentyl)hypoxanthine (3f). Method D: Compound **2f**, 1.96 g (5.2 mmol); Dowex 1; 0-1 M CH_3COOH; crystallized from H_2O; obtained 1.2 g (79%) of compound **3f**. The data are consistent with the lit.[19]

4.1.9.28. 9-(5-phosphonopentyl)guanine (3 g). Method D: Compound **2g**, 1.02 g (2.6 mmol); applied on the column of Dowex (50 × 8). After elution from the column with 2.5% aqueous ammonia, evaporation and redissolving the residue in H_2O the resultant solution was acidified by1 M HCl. Solid was filtered; obtained 0.6 g (75%) of compound **3g**. ESI [M−H] 300.0 (100).^{1}H NMR (D_2O + NaOD): 7.73 (s, 1H, H-8); 4.03 (t, 2H, $J_{1'-2'}$ = 7.2 Hz, H-1′); 1.82 (m, 2H, H-2′); 1.52 (m, 2H, H-4′); 1.30-1.41 (m, 4H, H-3′, 5′). ^{13}C NMR (D_2O + NaOD): 166.09 (C-6); 159.42 (C-2); 151.92 (C-4); 139.68 (C-8); 117.62 (C-5); 44.13 (C-1′); 29.86 (d, $J_{5'-P}$ = 130.7 Hz, C-5′); 29.53 (C-2′); 28.40 (d, $J_{3'-P}$ = 17.6 Hz, C-3′); 24.30 (d, $J_{4'-P}$ = 3.8 Hz, C-3′). For $C_{10}H_{16}N_5O_4P$. 2/3 H_2O (301.24) calcd: C, 38.34; H, 5.58; N, 22.36; P, 9.89. Found: C, 38.50; H, 5.70; N, 22.33; P, 10.05.

4.1.9.29. 9-(6-Phosphonohexyl)hypoxanthine (3 h). Method D: Compound **2h**, 0.4 g (1.0 mmol); Dowex 1, 0-0.5 M HCOOH; crystalized from H_2O; obtained 0.26 g (81%) of compound **3h**. ESI [M+Na] 323.2 (50).^{1}H NMR (D_2O+NaOD): 8.15 (s, 1H, H-2); 8.08 (s, 1H, H-8); 4.21 (t, 2H, $J_{1''-2'}$ = 7.1 Hz, H-1′); 1.86 (m, 2H, H-2′); 1.45 (m, 2H, H-5′); 1.26-1.40 (m, 6H, H-3′, 4′, 6′). ^{13}C NMR (D_2O+NaOD): 161.26 (C-6); 149.60 (C-4); 147.72 (C-2); 142.56 (C-8); 123.80 (C-5); 44.95 (C-1′); 30.90 (d, $J_{4'-P}$ = 17.3 Hz, C-4′); 29.84 (C-2′); 29.77 (d, $J_{6'-P}$ = 130.7 Hz, C-6′); 26.08 (C-3′); 24.46 (d, $J_{5'-P}$ = 4.1 Hz, C-5′). For $C_{11}H_{17}N_4O_4P$. ½ H_2O (300.25) calcd: C, 42.72; H, 5.87; N, 18.12; P, 10.02. Found: C, 42.65; H, 5.84; N, 18.02; P, 10.04.

4.1.9.30. 9-(6-phosphonohexyl)guanine (3i). Method D: Compound **2i**, 0.5 g (1.3 mmol); Dowex 1;0.5-1 M AcOH; extracted with hot water from D1; obtained 0.08 g (20%) of compound **3i**; ESI[M−H] 314.1 (100). ^{1}H NMR (D_2O+NaOD): 7.71 (s, 1H, H-8); 4.03 (t, 2H, $J_{1'-2'}$ = 7.1 Hz, H-1′); 1.81 (m, 2H, H-2′); 1.47 (m, 2H, H-5′); 1.29-1.40 (m, 6H, H-3′, 4′, 6′). ^{13}C NMR (D_2O+NaOD): 168.80 (C-6); 161.61 (C-2); 151.93 (C-4); 139.22 (C-8); 118.12 (C-5); 44.11 (C-1′); 31.07 (d, $J_{4'-P}$ = 17.3 Hz, C-4′); 29.99 (d, $J_{6'-P}$ = 130.5 Hz, C-6′); 29.76 (C-2′); 26.20 (C-3′); 24.62 (d, $J_{5'-P}$ = 4.2 Hz, C-5′). For $C_{11}H_{18}N_5O_4P$. H_2O (315.26) calcd: C, 39.64; H, 6.05; N, 21.01; P, 9.29. Found: C, 39.34; H, 5.74; N, 20.80; P, 9.60.

4.1.9.31. 9-(7-Phosphonoheptyl)hypoxanthine (3j). Method D: Compound **2j**, 0.91 g (2.3 mmol); Dowex 1, 0-1 M HCOOH; crystalized from H_2O; obtained 0.5 g (70%) of compound **3j**. ESI [M+Na] 313.2 (100).^{1}H NMR (D_2O): 8.13 (s, 1H, H-2); 7.98 (s, 1H, H-8); 4.18 (t, 2H, $J_{1'-2'}$ = 7.0 Hz, H-1′); 1.85 (m, 2H, H-2′); 1.45 (m, 2H, H-6′); 1.24-1.40 (m, 8H, H-3′, 4′, 5′, 7′). ^{13}C NMR (D_2O): 166.08 (C-6); 152.98 (C-2); 150.24 (C-4); 141.49 (C-8); 123.73 (C-5); 44.62 (C-1′); 31.46 (d, $J_{5'-P}$ = 17.3 Hz, C-5′); 30.03 (d, $J_{7'-P}$ = 130.8 Hz, C-7′); 29.82 (C-2′); 28.69 (C-4′); 26.45 (C-3′); 24.72 (d, *J*

$J_{6'\text{-P}}$ = 4.0 Hz, C-6′). For $C_{12}H_{19}N_4O_4P$. 1/3 H_2O (314.28) calcd: C, 45.00; H, 6.19; N, 17.49. Found: C, 45.31; H, 6.22; N, 17.47.

4.1.9.32. 9-[7-(phosphonoheptyl]guanine (3 k). Method D: Compound **2k**, 1.36 g (3.2 mmol); Dowex 1; 0.5-1 M AcOH; a suspension was made on the column; eluted by 0.7 M HCl; neutralized applied on Dowex (50 × 8) and eluted with 2.5% aqueous ammonia. The solvent was evaporated and the residue redissolved in H_2O, acidified by HCl and filtered; obtained 0.55 g (51%) of compound **3k**. The data were consistent with literature.[20]

4.1.9.33. 2-Amino-6-chloro-9-[6-(phosphonohexyl]-9*H*-purine (4a). Method A: compound **1i**, 0.39 g (0.96 mmol); HPLC; obtained 0.17 g (55%) of compound **4a**. ESI [M−H] 332.0 (100).^{1}H NMR (D_2O): 8.04 (s, 1H, H-8); 4.05 (t, 2H, $J_{1'\text{-}2'}$ = 7.3 Hz, H-1′); 1.79 (m, 2H, H-2′); 1.44–1.54 (m, 4H, H-5′,6′); 1.37 (m, 2H, H-4′); 1.23 (m, 2H, H-3′). ^{13}C NMR (D_2O): 159.66 (C-2); 153.61 (C-4); 150.70 (C-6); 145.15 (C-8); 124.20 (C-5); 44.56 (C-1′); 30.42 (d, $J_{4'\text{-P}}$ = 17.2 Hz, C-4′); 29.20 (C-2′); 28.73 (d, $J_{6'\text{-P}}$ = 132.6 Hz, C-6′); 26.00 (C-3′); 23.79 (d, $J_{5'\text{-P}}$ = 4.3 Hz, C-5′). For $C_{11}H_{17}ClN_5O_3P$ (333.71) calcd: C, 39.59; H, 5.11; N, 20.99. Found: C, 39.21; H, 5.11; N, 20.69.

4.1.9.34. 6-Chloro-9-[7-(phosphonoheptyl]-9*H*-purine (4b). Method A: compound **1j**, 0.53 g (1.4 mmol); HPLC; obtained 0.20 g (48%) of compound **4b**. ESI [M−H] 331.2 (100).^{1}H NMR (D_2O): 8.66 (s, 1H, H-2); 8.53 (s, 1H, H-8); 4.32 (t, 2H, $J_{1'\text{-}2'}$ = 7.2 Hz, H-1′); 1.89 (m, 2H, H-2′); 1.38–1.50 (m, 4H, H-6′,7′); 1.25–1.36 (m, 6H, H-3′,4′,5′). ^{13}C NMR (D_2O): 151.88 (C-4); 151.78 (C-2); 150.33 (C-6); 148.45 (C-8); 131.29 (C-5); 45.35 (C-1′); 31.11 (d, $J_{5'\text{-P}}$ = 17.2 Hz, C-5′); 29.45 (C-2′); 29.44 (d, $J_{7'\text{-P}}$ = 131.6 Hz, C-7′); 28.49 (C-4′); 26.33 (C-3′); 24.27 (d, $J_{6'\text{-P}}$ = 4.2 Hz, C-6′). For $C_{12}H_{18}ClN_4O_3P$. 1/2 H_2O (332.72) calcd: C, 42.55; H, 5.55; N, 16.54. Found: C, 42.49; H, 5.34; N, 16.29.

4.1.9.35. 2-Amino-6-chloro-9-[7-(phosphonoheptyl]-9*H*-purine (4c). Method A: compound **1k**, 0.6 g (1.3 mmol); HPLC; obtained 0.26 g (54%) of compound **4c**. ESI [M−H] 346.2 (100).^{1}H NMR (D_2O): 8.05 (s, 1H, H-8); 4.05 (t, 2H, $J_{1'\text{-}2'}$ = 7.2 Hz, H-1′); 1.79 (m, 2H, H-2′); 1.24–1.50 (m, 10H, H-3′, 7′). ^{13}C NMR (D_2O): 159.67 (C-2); 153.63 (C-4); 150.70 (C-6); 145.20 (C-8); 124.21 (C-5); 44.62 (C-1′); 31.44 (d, $J_{5'\text{-P}}$ = 17.4 Hz, C-5′); 30.03 (d, $J_{7'\text{-P}}$ = 130.6 Hz, C-7′); 29.33 (C-2′); 28.64 (C-4′); 26.41 (C-3′); 24.70 (d, $J_{6'\text{-P}}$ = 4.0 Hz, C-6′). For$C_{12}H_{19}ClN_5O_3P$ (347.74) calcd: C, 41.45; H, 5.51; N, 20.14. Found: C, 41.16; H, 5.51; N, 19.94.

4.1.9.36. 6-Bromo-9-(3-phosphonopropyl)-9*H*-purine (5a). Method H: Compound **1b**, 1 g (2.7 mmol); crystallized H_2O; obtained 0.6 g (78%) of compound **5a**. ESI [M−H] 319.0 (100).^{1}H NMR (D_2O+NaOD): 8.62 (s, 1H, H-2); 8.59 (s, 1H, H-8); 4.40 (t, 2H, $J_{1'\text{-}2'}$ = 7.2 Hz, H-1′); 2.13 (m, 2H, H-2′); 1.56 (m, 2H, H-3′). ^{13}C NMR (D_2O+NaOD): 151.83 (C-2); 150.61 (C-4); 148.35 (C-8); 142.31 (C-6); 134.01 (C-5); 46.02 (d, $J_{1'\text{-P}}$ = 19.3 Hz, C-1′); 25.77 (d, $J_{3'\text{-P}}$ = 133.7 Hz, C-3′); 24.60 (d, $J_{2'\text{-P}}$ = 3.5 Hz, C-1′). For $C_8H_{10}BrN_4O_3P$ (321.07) calcd: C, 29.93; H, 3.14; N, 17.45; P, 9.65. Found: C, 29.67; H, 3.12; N, 17.08; P, 9.59.

4.1.9.37. 2-Amino-6-bromo-9-(3-phosphonopropyl)-9*H*-purine (5b). Method H: Compound **1c**, 0.75 g (1.9 mmol); HPLC; obtained 0.24 g (36%) of compound **5b**. ESI [M−H] 334.0 (100).^{1}H NMR (D_2O+NaOD): 8.14 (s, 1H, H-8); 4.13 (t, 2H, $J_{1'\text{-}2'}$ = 7.3 Hz, H-1′); 2.02 (m, 2H, H-2′); 1.43 (m, 2H, H-3′). ^{13}C NMR (D_2O+NaOD): 159.64 (C-2); 152.48 (C-4); 145.30 (C-8); 142.79 (C-6); 127.03 (C-5); 45.74 (d, $J_{1'\text{-P}}$ = 19.2 Hz, C-1′); 26.53 (d, $J_{3'\text{-P}}$ = 130.1 Hz, C-3′); 24.98 (C-2′). For $C_8H_{11}BrN_5O_3P$. 1/2 H_2O (336.08) calcd: C, 27.84; H, 3.50; N, 20.29; P, 8.98. Found: C, 28.17; H, 3.82; N, 19.97; P, 9.00.

4.1.9.38. 6-Bromo-9-(5-phosphonopentyl)-9*H*-purine (5c). Method H: Compound **1f**, 1 g (2.5 mmol); 20 min. after neutralization a precipitate was isolated; the filtrate was concentrated and purified by HPLC and both portions were crystallized from H_2O; obtained 0.56 g (71%) of compound **5c**. ESI [M−H] 348.9 (100).^{1}H NMR (D_2O): 8.60 (s, 1H, H-2); 8.56 (s, 1H, H-8); 4.34 (t, 2H, $J_{1'\text{-}2'}$ = 7.1 Hz, H-1′); 1.93 (m, 2H, H-2′); 1.55 (m, 2H, H-4′); 1.45 (m, 2H, H-5′); 1.36 (m, 2H, H-3′). ^{13}C NMR (D_2O): 151.71 (C-2); 150.56 (C-4); 148.37 (C-8); 142.23 (C-6); 133.95 (C-5); 45.33 (C-1′); 29.22 (C-2′); 29.06 (d, $J_{5'\text{-P}}$ = 131.6 Hz, C-5′); 28.11 (d, $J_{3'\text{-P}}$ = 17.9 Hz, C-3′); 23.77 (d, $J_{4'\text{-P}}$ = 3.2 Hz, C-4′). For $C_{10}H_{14}BrN_4O_3P$ (349.12) calcd: C, 34.40; H, 4.04; N, 16.05; P, 8.87. Found: C, 34.34; H, 3.88; N, 15.71; P, 9.02.

4.1.9.39. 2-Amino-6-bromo-9-(5-phosphonopentyl)]-9*H*-purine (5d). Method H: Compound **1g**, 1.29 g (3.2 mmol); HPLC; obtained 0.5 g (43%) of compound **5d**. ESI [M−H] 362.0 (100).^{1}H NMR (D_2O): 8.11 (s, 1H, H-8); 4.09 (t, 2H, $J_{1'\text{-}2'}$ = 7.2 Hz, H-1′); 1.84 (m, 2H, H-2′); 1.51 (m, 2H, H-4′); 1.29–1.39 (m, 4H, H-3′, 5′). ^{13}C NMR (D_2O):159.64 (C-2); 152.49 (C-4); 145.30 (C-8); 142.78 (C-6); 127.00 (C-5); 44.66 (C-1′); 29.84 (d, $J_{5'\text{-P}}$ = 130.7 Hz, C-5′); 29.12 (C-2′); 28.39 (d, $J_{3'\text{-P}}$ = 17.7 Hz, C-3′); 24.25 (d, $J_{4'\text{-P}}$ = 3.6 Hz, C-4′). For $C_{10}H_{15}BrN_5O_3P$ (364.13) calcd: C, 32.98; H, 4.15; N, 19.23; P, 8.51. Found: C, 32.78; H, 3.96; N, 18.85; P, 8.71.

4.1.9.40. 6-Bromo-9-(6-phosphonohexyl)-9*H*-purine (5e). Method H: Compound **1h**, 0.5 g (1.2 mmol); HPLC; obtained 0.17 g (38%) of compound **5e**. ESI [M−H] 361.2 (100).^{1}H NMR (D_2O+NaOD): 8.62 (s, 1H, H-2); 8.56 (s, 1H, H-8); 4.33 (t, 2H, $J_{1'\text{-}2'}$ = 7.2 Hz, H-1′); 1.92 (m, 2H, H-2′); 1.45 (m, 2H, H-5′); 1.29–1.40 (m, 6H, H-3′, 4′, 6′). ^{13}C NMR (D_2O+NaOD): 151.76 (C-2); 150.65 (C-4); 148.41 (C-8); 142.27 (C-6); 134.01 (C-5); 45.42 (C-1′); 30.92 (d, $J_{4'\text{-P}}$ = 17.3 Hz, C-4′); 29.92 (d, $J_{6'\text{-P}}$ = 130.6 Hz, C-6′); 29.44 (C-2′); 26.16 (C-3′); 24.55 (d, $J_{5'\text{-P}}$ = 4.0 Hz, C-5′). For $C_{11}H_{16}BrN_4O_3P$ (363.15) calcd: C, 36.38; H, 4.44; N, 15.43; P, 8.53. Found: C, 36.23; H, 4.46; N, 15.22; P, 8.82.

4.1.9.41. 2-Amino-6-bromo-9-(6-phosphonohexyl)-9*H*-purine (5f).Method H: Compound **1i**, 0.5 g (1.2 mmol); HPLC; obtained 0.20 g (44%) of compound **5f**. ESI [M+Na] 400.1 (85).^{1}H NMR (D_2O+NaOD): 8.11 (s, 1H, H-8); 4.10 (t, 2H, $J_{1'\text{-}2'}$ = 7.1 Hz, H-1′); 1.83 (m, 2H, H-2′); 1.45 (m, 2H, H-5′); 1.27–1.38 (m, 6H, H-3′, 4′, 6′). ^{13}C NMR (D_2O+NaOD): 159.73 (C-2); 152.63 (C-4); 145.40 (C-8); 142.88 (C-6); 127.14 (C-5); 44.69 (C-1′); 30.94 (d, $J_{4'\text{-P}}$ = 17.2 Hz, C-4′); 29.93 (d, $J_{6'\text{-P}}$ = 130.9 Hz, C-6′); 29.27 (C-2′); 26.12 (C-3′); 24.55 (d, $J_{5'\text{-P}}$ = 4.0 Hz, C-5′). For $C_{11}H_{17}BrN_5O_3P$. ½ H_2O (378.16) calcd: C, 34.12; H, 4.69; N, 18.09; P, 8.00. Found: C, 34.27; H, 4.60; N, 17.75; P, 8.30.

4.1.9.42. Diisopropyl 9-(3-phosphonopropyl)]xanthine (6a). Method E: Compound **2c**, 1.5 g (4.2 mmol); obtained 1.43 g (95%) of compound **6a**. ESI [M−H] 357.1 (100).^{1}H NMR (DMSO-d_6): 7.69 (s, 1H, H-8); 4.52 (dn, 2H, $J_{\text{H–C–O–P}}$ = 8.0 Hz, $J_{\text{CH–CH3}}$ = 6.2 Hz, CH-*i*Pr); 4.08 (t, 2H, $J_{1'\text{-}2'}$ = 6.9 Hz, H-1″); 1.81 (m, 2H, H-2′); 1.62 (m, 2H, H-3′); 1.22 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 158.23 (C-6); 151.21 (C-2); 140.68 (C-4); 137.21 (C-8); 115.75 (C-5); 69.67 (d, $J_{\text{C–O–P}}$ = 6.4 Hz, CH-*i*Pr); 44.19 (d, $J_{1'\text{-P}}$ = 21.4 Hz, C-1′); 23.94 (d, $J_{\text{C–O–P}}$ = 4.2 Hz CH_3-*i*Pr); 23.68 (d, $J_{2'\text{-P}}$ = 4.0 Hz, C-2′); 23.03 (d, $J_{3'\text{-P}}$ = 142.3 Hz, C-3′).

4.1.9.43. Diisopropyl 9-(4-phosphonobutyl)xanthine (6b). Method E: Compound **2e**, 2.07 g (5.6 mmol); crystallized from $CHCl_3$ acetone; obtained 1.6 g (77%) compound **6b**. ESI [M+Na] 395.1 (100).^{1}H NMR (DMSO-d_6):10.72 (br s, 1H, NH); 7.66 (s, 1H, H-8); 4.51 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 4.01 (t, 2H, $J_{1'-2'}$ = 7.0 Hz, H-1′);1.65–1.76 (m, 4H, H-2′, 4′); 1.40 (m, 2H, H-3′);1.21 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.19 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr).13C NMR (DMSO-d_6):158.25 (C-6); 151.44 (C-2); 140.99 (C-4); 137.13 (C-8); 115.63 (C-5); 69.33 (d, J_{C-O-P} = 6.5 Hz, CH-*i*Pr); 43.42 (C-1′); 30.44 (d, $J_{2'-P}$ = 6.8 Hz, C-2′); 25.36 (d, $J_{4'-P}$ = 140.7 Hz, C-4′); 23.99 (m, CH_3-*i*Pr); 19.22 (d, $J_{3'-P}$ = 4.8 Hz, C-3′). For $C_{15}H_{25}N_4O_5P$. 1/3 H_2O (372.36) calcd: C, 47.62; H, 6.84; N, 14.81. Found: C, 47.68; H, 6.82; N, 14.85; P.

4.1.9.44. Diisopropyl 9-(5-phosphonopentyl)xanthine (6c). Method E: Compound **2g**, 3.5 g (9.1 mmol); obtained 3.3 g (94%) of compound **6c**. ESI [M+Na] 409.10 (100).^{1}H NMR (DMSO-d_6): 10.66 (br s, 1H, NH); 7.66 (s, 1H, H-8); 4.51 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.97 (t, 2H, $J_{1'-2'}$ = 7.3 Hz, H-1′); 1.65 (m, 2H, H-2′); 1.62 (m, 2H, H-5′); 1.45 (m, 2H, H-4′); 1.30 (m, 2H, H-3′); 1.21 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 158.35 (C-6); 141.46 (C-4); 137.07 (C-8); 115.58 (C-5); 69.27 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 43.85 (C-1′); 29.43 (C-2′); 26.82 (d, $J_{3'-P}$ = 16.2 Hz, C-3′); 25.96 (d, $J_{5'-P}$ = 139.9 Hz, C-5′); 24.03 (d, $J_{C-C-O-P}$ = 4.3 Hz, CH_3-*i*Pr); 22.13 (d, $J_{4'-P}$ = 5.2 Hz, C-4′).

4.1.9.45. Diisopropyl 9-(6-phosphonohexyl)xanthine (6d). Method E: Compound **2i**, 0.86 g (2.1 mmol); obtained 0.81 g (94%) of compound **6d**. Compound was used in the next step

4.1.9.46. 9-(3-Phosphonopropyl)]xanthine (7a). Method D: Compound **6a**, 4.5 g (12.6 mmol); crystallized from H_2O; obtained 2.4 g (70%) of compound **7a**. ESI [M+H] 275.0 (100).^{1}H NMR (D_2O+NaOD): 7.66 (s, 1H, H-8); 4.04 (t, 2H, $J_{1'-2'}$ = 7.1 Hz, H-1′); 2.03 (m, 2H, H-2′); 1.58 (m, 2H, H-3′). ^{13}C NMR (DMSO-d_6): 160.68 (C-6); 154.69 (C-2); 144.74 (C-4); 139.71 (C-8); 115.72 (C-5); 45.56 (d, $J_{1'-P}$ = 16.8 Hz, C-1′); 25.10 (d, $J_{3'-P}$ = 131.2 Hz, C-3′); 24.55 (C-2′). For $C_8H_{11}N_4O_5P$ (274.17). H_2O calcd: C, 32.89; H, 4.48; N, 19.15; P, 10.60. Found: C, 32.73; H, 4.41; N, 18.85; P, 10.97.

4.1.9.47. 9-(4-Phosphonobutyl)xanthine (7b). Method D: Compound **6b**, 1.56 g (4.2 mmol); crystallized from H_2O; obtained 0.725 g (60%) of compound **7b**. ESI [M+H] 288.3 (100).^{1}H NMR (D_2O): 7.73 (s, 1H, H-8); 4.03 (t, 2H, $J_{1'-2'}$ = 7.2 Hz, H-1′); 1.87 (q, 2H, $J_{2'-1'}$ = $J_{1'-2'}$ = 7.1 Hz, H-2′); 1.49–1.64 (m, 4H, 3′, 4′). ^{13}C NMR (D_2O): 161.40 (C-6); 151.36 (C-4); 139.96 (C-8); 115.40 (C-5); 44.00 (C-1′); 30.89 (d, $J_{3'-P}$ = 16.7 Hz, C-3′); 28.02 (d, $J_{4'-P}$ = 132.8 Hz, C-4′); 21.00 (C-2′). For $C_9H_{13}N_4O_5P$. 1/3 H_2O (288.2) calcd: C, 36.74; H, 4.68; N, 19.04; P, 10.53. Found: C, 36.71; H, 4.44; N, 18.71; P, 10.93.

4.1.9.48. 8-Bromo-9-(5-phosphonopentyl)]xanthine (7c). Method D: Compound **6c**, 3.3 g (8.5 mmol); 8-position of the base was unexpectedly brominated during the reaction. Dowex 1 0–0.8 M HCOOH; crystallized from H_2O; obtained 1.6 g (49%) of compound **7c**. ESI [M−H] 379.0/381 (100).^{1}H NMR (D_2O): 3.99 (t, 2H, $J_{1'-2'}$ = 7.4 Hz, H-1′); 1.77 (m, 2H, H-2′); 1.52 (m, 2H, H-4′); 1.33–1.44 (m, 4H, H-3′, 5′). ^{13}C NMR (D_2O): 160.59 and 160.13 (C-2 and C-6); 155.11 (C-4); 124.32 (C-8); 115.70 (C-5); 44.71 (C-1′); 29.69 (d, $J_{5'-P}$ = 131.1 Hz, C-5′); 29.00 (C-2′); 28.35 (d, $J_{3'-P}$ = 17.4 Hz, C-3′); 24.33 (C-4′). For $C_{10}H_{14}BrN_4O_5P$. 2/3 H_2O (381.12) calcd: C, 30.55; H, 3.93; N, 14.25. Found: C, 30.45; H, 3.60; N, 13.98.

4.1.9.49. 9-(6-Phosphonohexyl)xanthine (7d). Method D: Compound **6d**, 0.8 g (2.0 mmol); crystallized from H_2O; obtained 0.27 g (43%) of compound **7d**. ESI [M−H] 315.2 (100). ^{1}H NMR (D_2O): 7.72 (s, 1H, H-8); 4.39 (t, 2H, $J_{1'-2'}$ = 7.1 Hz, H-1′); 1.80 (p, 2H, $J_{2'-1'}$ = $J_{2'-3'}$ = 7.1 Hz, H-2′); 1.46 (m, 2H, H-5′); 1.28–1.39 (m, 6H, H-3′, 4′, 6′). ^{13}C NMR (D_2O): 161.74 (C-6); 160.42 (C-2); 154.12 (C-4); 140.20 (C-8); 115.27 (C-5); 44.06 (C-1′); 31.09 (d, $J_{4'-P}$ = 17.6 Hz, C-4′); 29.98 (d, $J_{6'-P}$ = 131.0 Hz, C-6′); 29.59 (C-2′); 26.18 (C-3′); 24.65 (d, $J_{5'-P}$ = 4.0 Hz, C-5′). For $C_{11}H_{17}N_4O_5P$ (316.25) calcd: C, 41.78; H, 5.42; N, 17.12. Found: C, 41.55; H, 5.40; N, 17.27.

4.1.9.50. 9-[7-(Phosphonoheptyl]xanthine (7e). Method E: Compound **3k**, 0.3 g (0.9 mmol); Dowex 1 HCOOH 1 M; crystallized from H_2O; obtained 0.18 g (60%) compound **7e**. ESI [M+Na] 329.2 (100).^{1}H NMR (D_2O): 7.75 (s, 1H, H-8); 4.07 (t, 2H, $J_{1'-2'}$ = 7.1 Hz, H-1′); 1.78 (m, 2H, H-2′); 1.45–1.60 (m, 4H, H-6′,7′); 1.24–1.36 (m, 6H, H-3′, 4′, 5′). ^{13}C NMR (D_2O): 160.35 (C-6); 152.99 (C-2); 141.84 (C-4); 139.62 (C-8); 115.75 (C-5); 45.69 (C-1′); 30.55 (d, $J_{5'-P}$ = 16.9 Hz, C-5′); 29.34 (C-2′); 28.35 (C-4′); 28.33 (d, $J_{7'-P}$ = 133.1 Hz, C-7′); 25.96 (C-3′); 23.52 (d, $J_{6'-P}$ = 4.6 Hz, C-6′). For $C_{12}H_{19}N_4O_5P$. 1/3 H_2O (330.28) calcd: C, 42.86; H, 5.89; N, 16.66. Found: C, 42.93; H, 5.72; N, 16.59.

4.1.9.51. Diethyl 2-amino-6-chloro-9-(2-phosphonoethyl)-9*H*-purine (9). 2-Amino-6-chloropurine 2 g (11.8 mmol) and caesium carbonate 384 mg (3.5 mmol) was suspended in 150 mL DMF. Diethyl vinylphosphonate 2.02 mL (13 mmol) was added and the mixture was stirred at 90 °C for 8 h. Solvent was evaporated in vacuo and the residue purified by column chromatography 5–10% MeOH in $CHCl_3$; obtained 932 mg (24%) of compound **9**. ESI [M+H] 334.1 (100).^{1}H NMR ($CDCl_3$): 7.83 (s, 1H, H-8); 5.23 (bs, 2H, NH_2); 4.37 (m, 2H, H-2′); 4.06 (m, 4H, CH_2-Et); 2.37 (dm, 2H, $J_{(H-C-P)}$ = 18.3 Hz, H-1′); 1.26 (t, 6H, $J_{(CH3-CH)}$ = 7.0 Hz, CH_3-Et); ^{13}C NMR ($CDCl_3$): 158.94 (C-2); 153.56 (C-4); 151.29 (C-6); 142.56 (C-8); 125.24 (C-5); 62.10 (d, $J_{(C-O-P)}$ = 6.6 Hz, CH_2-Et); 38.59 (d, $J_{(C-C-P)}$ = 3.4 Hz, C-2′); 26.05(d, J_{C-P} = 141.4 Hz, C-1′); 16.31 (d, $J_{(C-C-O-P)}$ = 6.1 Hz, CH_3-Et). For $C_{11}H_{17}ClN_5O_3P$ (333.71) calcd: C, 39.59; H, 5.13; N, 20.99; P, 9.28. Found: C, 39.38; H, 4.94; N, 20.75; P, 9.16

4.1.9.52. 9-(2-phosphonoethyl)guanine (10). From compound **9**, 932 mg (2.8 mmol) by Method C: without purification used for the next step Method D: crystallized H_2O/MeOH; obtained 355 mg (49%) of compound **10**. ESI [M−H] 258.0 (100).^{1}H NMR (D_2O): 7.77 (s, 1H, H-8); 4.19 (d, 2H, H-2′); 1.97 (d, 2H, H-1′); ^{13}C NMR (D_2O): 168.89 (C-6); 161.67 (C-2); 151.76 (d, C-4); 138.82 (C-8); 118.18 (C-5); 41.04 (C-2′); 31.22 (d, $J_{(C-P)}$ = 124.7 Hz, C-1′); For $C_7H_{10}N_5O_4P$ (259.16) calcd: C, 32.44; H, 3.89; N, 27.02; P, 11.95. Found: C, 32.36; H, 4.02; N, 27.13; P, 11.78.

4.1.9.53. 9-(1-phosphonomethyl)hypoxanthine (12a). Method E2: Compound **11a**, 300 mg (1.3 mmol); crystallized H_2O/MeOH; obtained 105 mg (35%) of compound **12a**. ESI [M+Na^+] 253.0 (100).^{1}H NMR (D_2O): 8.17 (s, 1H, H-8); 8.14 (s, 1H, H-2), 4.31 (d, 2H, $J_{(H-C-P)}$ = 12.0 Hz, H-1′); ^{13}C NMR (D_2O): 168.35 (C-6); 153.93 (C-2); 150.82 (d, $J_{(C4-P)}$ = 3.8 Hz), C-4); 141.35 (d, $J_{(C8-P)}$ = 1.1 Hz, C-8); 123.10 (C-5); 42.77 (d, $J_{(C-P)}$ = 133.9 Hz, C-1′); For $C_6H_7N_4O_4P$ (230.12) calcd: C, 31.32; H, 3.07; N, 24.35; P, 13.46. Found: C, 31.27; H, 3.32; N, 24.21; P, 13.26.

4.1.9.54. 9-(2-phosphonoethyl)hypoxanthine (12b). Method E2: Compound **11b**, 300 mg (1.24 mmol); crystallized H_2O/MeOH; obtained 130 mg (43%) of compound **12b**. ESI [M−H] 243.0 (100).^{1}H NMR (D_2O): 8.77 (s, 1H, H-8); 8.30 (s, 1H, H-2); 4.58 (dm, 2H, $J_{(H-C-C-P)}$ = 12.5 Hz, H-2′); 2.32 (dm, 2H, $J_{(H-C-P)}$ = 17.7 Hz, H-1′);

^{13}C NMR (D_2O): 156.91 (C-6); 148.51 (C-4); 148.11 (C-2); 141.52 (C-8); 119.80 (C-5); 41.62 (C-2′); 28.32 (d, $J_{(C-P)}$ = 133.0 Hz, C-1′); For $C_7H_8N_4O_4P$ (244.14) calcd: C, 34.44; H, 3.72; N, 22.95; P, 12.69. Found: C, 34.28; H, 4.01; N, 21.99; P, 12.54.

4.1.9.55. Diisopropyl 6-chloro-9-[1-(phosphonoethoxy)methyl]-9*H*-purine (14a). Method G: 6-Chloropurine 1.18 g (7.6 mmol); diisopropyl 2-(chloromethoxy)ethylphosphonate (**13a**)[20]; room temperature 3 h, column chromatography 0-3% MeOH in $CHCl_3$; obtained 0.91 g (32%) of compound **14a**. ESI [M+H] 399.0 (100).^{1}H NMR (DMSO-d_6):8.86 (s, 1H, H-8); 8.84 (s, 1H, H-2); 5.71 (s, 2H, H-1′); 4.48 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.68 (dt, 2H, $J_{3'-P}$ = 13.0 Hz, $J_{3'-4'}$ = 7.3 Hz, H-3′); 2.00 (dt, 2H, $J_{4'-P}$ = 18.4 Hz, $J_{4'-3'}$ = 7.3 Hz, H-4′); 1.16 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.13 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6):152.40 (C-4); 152.32 (C-2); 149.55 (C-6); 148.02 (C-8); 131.10 (C-5); 72.80 (C-1′); 69.72 (d, CH-*i*Pr); 63.83 (d, $J_{3'-P}$ = 1.1 Hz, C-3′); 27.04 (d, $J_{4'-P}$ = 139.1 Hz, C-4′); 23.96 (d, $J_{C-C-O-P}$ = 3.7 Hz, CH_3-*i*Pr); 23.85 (d, $J_{C-C-O-P}$ = 4.7 Hz, CH_3-*i*Pr).

4.1.9.56. Diisopropyl 2-amino-6-chloro-9-[1-(phosphonoethoxy)methyl]-9*H*-purine (14b). Method G: 2-Amino-6-chloropurine 2.5 g (14.7 mmol); diisopropyl 2-(chloromethoxy) ethylphosphonate (**13a**)[23]; room temperature 36 h; column chromatography 0-3% MeOH in $CHCl_3$; obtained 2.17 g (38%) of compound **14b**. ESI [M+Na] 414.0 (100).^{1}H NMR (DMSO-d_6): 8.27 (s, 1H, H-8); 7.02 (br s, 2H, NH_2); 5.44 (s, 2H, H-1′); 4.49 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.61 (dt, 2H, $J_{3'-P}$ = 12.7 Hz, $J_{3'-4'}$ = 7.3 Hz, H-3′); 1.98 (dt, 2H, $J_{4'-P}$ = 18.4 Hz, $J_{4'-3'}$ = 7.3 Hz, H-4′); 1.18 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.15 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 160.36 (C-2); 154.54 (C-4); 149.83 (C-6); 143.58 (C-8); 123.39 (C-5); 72.02 (C-1′); 69.74 (d, $J_{C-C-O-P}$ = 6.3 Hz, CH-*i*Pr); 63.19 (C-3′); 27.06 (d, $J_{4'-P}$ = 139.1 Hz, C-4′); 23.97 (d, $J_{C-C-O-P}$ = 3.8 Hz, CH_3-*i*Pr); 23.87 (d, $J_{C-C-O-P}$ = 4.7 Hz, CH_3-*i*Pr).

4.1.9.57. Diisopropyl 6-chloro-9-[1-(phosphonopropoxy) methyl]-9*H*-purine (14c). Method G: 6-Chloropurine 1 g (6.5 mmol); diisopropyl 3-(chloromethoxy)propylphosphonate (**13b**)[23]; room temperature 12 h; column chromatography 0-5% MeOH in $CHCl_3$; obtained 1.37 g (54%) of compound **14c**. ESI [M+Na] 413.1 (10).^{1}H NMR (DMSO-d_6): 8.86 (s, 1H, H-8); 8.83 (s, 1H, H-2); 5.69 (s, 2H, H-1′); 4.53 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.53 (m, 2H, H-3′); 1.54-1.72 (m, 4H, H-4′,5′); 1.17 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.15 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 152.38 (C-4); 152.27 (C-2); 149.48 (C-6); 147.93 (C-8); 131.08 (C-5); 73.16 (C-1′); 69.32 (d, J_{C-O-P} = 6.3 Hz, CH-*i*Pr); 68.93 (d, $J_{3'-P}$ = 7.1 Hz, C-3′); 23.92 (m, CH_3-*i*Pr); 22.79 (d, $J_{4'-P}$ = 4.8 Hz, C-4′); 22.53 (d, $J_{5'-P}$ = 142.0 Hz, C-5′).

4.1.9.58. Diisopropyl 2-amino-6-chloro-9-[1-(phosphonopropoxy)methyl]-9*H*-purine (14d). Method G: 2-Amino-6-chloropurine 3 g (18 mmol); diisopropyl 3-(chloromethoxy)propylphosphonate (**13b**)[23]; room temperature 12 h; column chromatography 0-4% MeOH in $CHCl_3$; obtained 3.5 g (49%) of compound **14d**. ESI [M+Na] 428.1 (100).^{1}H NMR (DMSO-d_6):8.26 (s, 1H, H-8); 7.03 (br s, 2H, NH_2); 5.43 (m, 2H, H-1′); 4.47 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.49 (m, 2H, H-3′); 1.54-1.71 (m, 4H, H-4′,5′); 1.18 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.17 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 160.32 (C-2); 154.50 (C-4); 149.76 (C-6); 143.51 (C-8); 123.43 (C-5); 72.45 (C-1′); 69.43 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 68.53 (d, $J_{3'-P}$ = 17.3 Hz, C-3′); 23.96 (m, CH_3-*i*Pr); 22.83 (d, $J_{4'-P}$ = 4.7 Hz, C-4′); 22.59 (d, $J_{5'-P}$ = 142.0 Hz, C-5′).

4.1.9.59. Diisopropyl 6-chloro-9-[1-(phosphonobutoxy) methyl]-9*H*-purine (14e).Method G: 6-Chloropurine 1 g (6.5 mmol); diisopropyl 4-(chloromethoxy)butylphosphonate (^{13}C)[20]; room temperature 5 h; column chromatography 0-2% MeOH in $CHCl_3$; obtained 1.43 g (55%) of compound **14e**. ESI [M+Na] 427.1 (100).^{1}H NMR (DMSO-d_6): 8.84 (s, 1H, H-8); 8.82 (s, 1H, H-2); 5.68 (s, 2H, H-1′); 4.47 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.52 (t, 2H, $J_{2'-3'}$ = 6.2 Hz, H-2′); 1.50–1.69 (m, 4H, H-4′,6′); 1.39 (m, 2H, H-5′); 1.19 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.16 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 152.33 (C-4); 152.23 (C-2); 149.48 (C-6); 147.89 (C-8); 131.07 (C-5); 73.17 (C-1′); 69.19 (d, J_{C-O-P} = 6.3 Hz, CH-*i*Pr); 68.68 (C-3′); 29.48 (d, $J_{4'-P}$ = 16.4 Hz, C-4′); 25.56 (d, $J_{6'-P}$ = 140.4 Hz, C-6′); 23.97 (m, CH_3-*i*Pr); 19.09 (d, $J_{5'-P}$ = 4.9 Hz, C-5′).

4.1.9.60. Diisopropyl 2-amino-6-chloro-9-[1-(phosphonobutoxy)methyl]-9*H*-purine (14f). Method G: 2-Amino-6-chloropurine 1.44 g (8.5 mmol); diisopropyl 4-(chloromethoxy) butylphosphonate (^{13}C)[23]; room temperature 24 h; column chromatography 0-2% MeOH in $CHCl_3$; obtained 1.85 g (52%) of compound **14f**. ESI [M+H] 420.0 (100).^{1}H NMR (DMSO-d_6): 8.25 (s, 1H, H-8); 6.99 (br s, 2H, NH_2); 5.42 (s, 2H, H-1′); 4.48 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.47 (t, 2H, $J_{3'-4'}$ = 6.2 Hz, H-3′); 1.50–1.62 (m, 4H, H-4′, 6′); 1.42 (m, 2H, H-5′); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.18 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 160.30 (C-2); 154.48 (C-4); 149.75 (C-6); 143.51 (C-8); 123.41 (C-5); 72.38 (C-1′); 69.22 (d, J_{C-O-P} = 6.3 Hz, CH-*i*Pr); 68.20 (C-3′); 29.52 (d, $J_{4'-P}$ = 16.3 Hz, C-4′); 25.57 (d, $J_{6'-P}$ = 140.7 Hz, C-6′); 23.97 (m, CH_3-*i*Pr); 19.14 (d, $J_{5'-P}$ = 4.7 Hz, C-5′).

4.1.9.61. Diisopropyl 9-[1-(phosphonoethoxy)methyl]hypoxanthine (15a). Method C: Compound **14a** 0.4 g (1.1 mmol); reflux 2 h; purified on HPLC; obtained 0.18 g (47%) of compound **15a**. ESI [M+Na] 381.0 (100).^{1}H NMR (DMSO-d_6): 8.21 (s, 1H, H-8); 8.09 (s, 1H, H-2); 5.53 (s, 2H, H-1′); 4.49 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.63 (dt, 2H, $J_{3'-P}$ = 12.3 Hz, $J_{3'-4'}$ = 7.4 Hz, H-3′); 1.99 (dt, 2H, $J_{4'-P}$ = 18.5 Hz, $J_{4'-3'}$ = 7.4 Hz, H-4′); 1.18 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.16 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 157.29 (C-6); 148.86 (C-4); 146.73 (C-2); 140.82 (C-8); 124.19 (C-5); 72.27 (C-1′); 69.74 (d, J_{C-O-P} = 6.3 Hz, CH-*i*Pr); 63.34 (C-3′); 27.05 (d, $J_{4'-P}$ = 139.0 Hz, C-4′); 23.99 (d, $J_{C-C-O-P}$ = 3.8 Hz, CH_3-*i*Pr); 23.88 (d, $J_{C-C-O-P}$ = 4.7 Hz, CH_3-*i*Pr).

4.1.9.62. Diisopropyl 9-[1-(phosphonoethoxy)methyl]guanine (15b). Method C: Compound **14b**, 1.5 g (3.8 mmol); reflux 2 h; column chromatography (MeOH: $CHCl_3$ 10:90); obtained 1.3 g (91%) of compound **15b**. ESI [M+Na] 396.1 (100).^{1}H NMR (DMSO-d_6): 10.67 (br s, 1H, NH); 7.82 (s, 1H, H-8); 6.52 (br s, 2H, NH_2); 5.33 (s, 2H, H-1′); 4.50 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.58 (dm, 2H, $J_{3'-P}$ = 12.0 Hz, H-3′); 1.98 (dm, 2H, $J_{4'-P}$ = 18.5 Hz, H-4′); 1.19 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.16 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 157.09 (C-6); 154.18 (C-2); 151.72 (C-4); 138.02 (C-8); 116.67 (C-5); 71.66 (C-1′); 69.78 (d, J_{C-O-P} = 8.3 Hz, CH-*i*Pr); 62.84 (C-3′); 27.08 (d, $J_{4'-P}$ = 138.9 Hz, C-4′); 24.01 (d, $J_{C-C-O-P}$ = 3.8 Hz, CH_3-*i*Pr); 23.90 (d, $J_{C-C-O-P}$ = 4.7 Hz, CH_3-*i*Pr).

4.1.9.63. Diisopropyl 9-[1-(phosphonopropoxy)methyl]hypoxanthine (15c). Method C: Compound **14c**, 0.8 g (2.1 mmol); reflux 2 h; partitioned between brine and $CHCl_3$; organic parts dried by $MgSO_4$; crystallized EtOAc petrolether; obtained 0.37 g (49%) of compound **15c**. ESI [M+Na] 395.1 (100).^{1}H NMR (DMSO-d_6): 12.41 (br s, 1H, NH); 8.23 (s, 1H, H-8); 8.08 (s, 1H, H-2); 5.52 (s, 2H, H-1′); 4.48 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.49 (m, 2H, H-3′); 1.54-1.64 (m, 4H, H-4′,5′); 1.19 (d, 6H,

$J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.17 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 156.87 (C-6); 148.77 (C-4); 146.33 (C-2); 140.87 (C-8); 124.19 (C-5); 72.67 (C-1′); 69.34 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 68.63 (d, $J_{3'-P}$ = 17.4 Hz, C-3′); 23.95 (m, CH_3-*i*Pr); 22.80 (d, $J_{4'-P}$ = 4.7 Hz, C-4′); 22.61 (d, $J_{5'-P}$ = 142.0 Hz, C-5′).

4.1.9.64. Diisopropyl 9-[1-(phosphonopropoxy)methyl]guanine (15d). Method C: Compound **14d**, 2 g (4.9 mmol); reflux 2 h; column chromatography (MeOH: $CHCl_3$ 5:95); obtained 1.32 g (69%) of compound **15d**. ESI [M+Na] 410.1 (100).^{1}H NMR (DMSO-d_6): 12.65 (br s, 1H, NH); 7.81 (s, 1H, H-8); 6.53 (br s, 2H, NH_2); 5.31 (s, 2H, H-1′); 4.49 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.46 (m, 2H, H-3′); 1.55-1.65 (m, 4H, H-4′,5′); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.18 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 157.03 (C-6); 154.11 (C-2); 151.64 (C-2); 137.92 (C-8); 116.72 (C-5); 72.08 (C-1′); 69.37 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 68.26 (d, $J_{3'-P}$ = 17.5 Hz, C-3′); 23.96 (m, CH_3-*i*Pr); 22.83 (d, $J_{4'-P}$ = 4.8 Hz, C-4′); 22.66 (d, $J_{5'-P}$ = 141.9 Hz, C-5′).

4.1.9.65. Diisopropyl-9-[1-(phosphonobutoxy)methyl]guanine (15f). Method C: Compound **14f**, 2 g (4.7 mmol); reflux 1 h; column chromatography (MeOH: $CHCl_3$ 10:90); obtained 1.32 g (69%) of compound **15f**. ESI [M−H] 400.1 (80).^{1}H NMR (DMSO-d_6): 10.65 (br s, 1H, NH); 7.81 (s, 1H, H-8); 6.52 (br s, 2H, NH_2); 5.30 (s, 2H, H-1′); 4.49 (dn, 2H, $J_{H-C-O-P}$ = 8.0 Hz, J_{CH-CH3} = 6.2 Hz, CH-*i*Pr); 3.43 (t, 2H, $J_{3'-4'}$ = 6.2 Hz, H-3′); 1.49-1.63 (m, 4H, H-4′, 6′); 1.43 (m, 2H, H-5′); 1.20 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr); 1.19 (d, 6H, $J_{CH3,CH}$ = 6.2 Hz, CH_3-*i*Pr). ^{13}C NMR (DMSO-d_6): 157.09 (C-6); 154.12 (C-2); 151.69 (C-4); 138.02 (C-8); 116.71 (C-5); 72.05 (C-1′); 69.31 (d, J_{C-O-P} = 6.4 Hz, CH-*i*Pr); 67.88 (C-3′);29.59 (d, $J_{4'-P}$ = 16.4 Hz, C-4′); 25.60 (d, $J_{6'-P}$ = 140.1 Hz, C-6′); 24.02 (m, CH_3-*i*Pr); 19.24 (d, $J_{5'-P}$ = 5.0 Hz, C-5′). For $C_{16}H_{28}N_5O_5P$. ½ MeOH (401.40) calcd: C, 47.48; H, 7.24; N, 16.78. Found: C, 47.46; H, 6.94; N, 16.94.

4.1.9.66. 9-[1-(Phosphonoethoxy)methyl]hypoxanine (16a). Method A: Compound **15a**, 0.15 g (0.4 mmol); obtained 0.07 g (61%) of compound **16a**. ESI [M−H] 272.9 (100). ^{1}H NMR (D_2O): 8.51 (s, 1H, H-8); 8.26 (s, 1H, H-2); 5.72 (s, 2H, H-1′); 3.83 (dt, 2H, $J_{3'-P}$ = 13.5 Hz, $J_{3'-4'}$ = 7.3 Hz, H-3′); 2.04 (dt, 2H, $J_{4'-P}$ = 18.1 Hz, $J_{4'-3'}$ = 7.3 Hz, H-4′). ^{13}C NMR (D_2O): 158.37 (C-6); 149.25 (C-4); 147.54 (C-2); 142.50 (C-8); 122.67 (C-5); 74.17 (C-1′); 65.12 (d, $J_{3'-P}$ = 1.4 Hz, C-3′); 28.32 (d, $J_{4'-P}$ = 133.6 Hz, C-4′). For $C_8H_{11}N_4O_5P$. H_2O (274.17) calcd: C, 32.89; H, 4.48; N, 19.18. Found: C, 32.72; H, 4.38; N, 18.82.

4.1.9.67. 9-[1-(phosphonoethoxy)methyl]guanine (16b). Method A: compound **15b**, 1.3 g (3.5 mmol); obtained 0.54 g (54%) of compound **16b**; The data were consistent with the literature[19]

4.1.9.68. 9-1-(Phosphonopropoxy)methyl]hypoxanthine (16c). Method A: Compound **15c**, 0.33 g (0.9 mmol); obtained 0.22 g (86%) of compound **16c**. ESI [M+H] 288.9 (100).^{1}H NMR (D_2O): 8.23 (s, 1H, H-8); 8.19 (s, 1H, H-2); 5.65 (s, 2H, H-1′); 3.60 (t, 2H, $J_{3'-4'}$ = 6.7 Hz, H-3′); 1.75 (m, 2H, H-4′); 1.35 (m, 2H, H-5′). ^{13}C NMR (D_2O): 161.16 (C-6); 149.74 (C-4); 148.40 (C-2); 142.58 (C-8); 124.07 (C-5); 76.77 (C-1′); 71.36 (d, $J_{3'-P}$ = 19.3 Hz, C-3′); 26.01 (d, $J_{5'-P}$ = 132.2 Hz, C-5′); 24.65 (d, $J_{4'-P}$ = 3.6 Hz, C-4′). For $C_9H_{13}N_4O_5P$. 1/4 H_2O (288.1) calcd: C, 36.93; H, 4.65; N, 19.14. Found: C, 36.95; H, 4.63; N, 19.04.

4.1.9.69. 9-[1-(phosphonopropoxy)methyl]guanine (16d). Method A: compound **15d**, 1.22 g (3.1 mmol);obtained 0.55 g (58%) of compound **16d**; the data were consistent with the liter ature.[23]

4.1.9.70. 9-[1-(Phosphonobutoxy)methyl]hypoxanthine (16e). Method C: Compound **17e** 0.86 g (2.7 mmol); 2 h room temperature; deionized on charcoal (applied on charcoal in batch adjusted with HCl to pH 3 and washed with water. The UV absorbing fraction was eluted by 2.5% aqueous ammonia.) Purified by HPLC; obtained 0.08 g (10%) of compound **16e**. ESI [M−H] 301.0 (100).^{1}H NMR (D_2O+NaOD): 8.29 (s, 1H) and 8.22 (s, 1H) H-2 and H-8; 5.67 (s, 2H, H-1′); 3.62 (t, 2H, $J_{3'-4'}$ = 6.4 Hz, H-3′); 1.62 (m, 2H, H-4′); 1.46-1.58 (m, 4H, H-5′, 6′). ^{13}C NMR (D_2O+NaOD): 159.13 (C-6); 149.50 (C-4); 146.93 (C-2); 142.83 (C-8); 124.02 (C-5); 73.93 (C-1′); 69.66 (C-3′); 30.08 (d, $J_{4'-P}$ = 16.8 Hz, C-4′); 27.48 (d, $J_{6'-P}$ = 133.9 Hz, C-6′); 19.98 (d, $J_{5'-P}$ = 4.4 Hz, C-5′). For $C_{10}H_{15}N_4O_5P$.2 H_2O (302.22) calcd: C, 35.51; H, 5.66; N, 16.56. Found: C, 35.45; H, 5.75; N, 16.78.

4.1.9.71. 9-[1-(phosphonobutoxy)methyl]guanine (16f). Method A: compound **15f**, 1.12 g (3.5 mmol); obtained 0.5 g (56%) of compound **16f**; the data were consistent with the literature.[20]

4.1.9.72. 6-Chloro-9-[1-(phosphonoethoxy)methyl]-9*H*-purine (17a). Method A: compound **14a**, 0.4 g (1.1 mmol); HPLC; obtained 0.13 g (42%) of compound **17a**. ESI [M−H] 290.8 (100).^{1}H NMR (D_2O): 8.77 (s, 1H, H-2); 8.71 (s, 1H, H-8); 5.79 (s, 2H, H-1′); 3.81 (m, 2H, H-3′); 1.91 (dm, 2H, $J_{4'-P}$ = 18.1 Hz, H-4′). ^{13}C NMR (D_2O): 152.63 (C-2); 152.19 (C-4); 150.86 (C-6); 148.37 (C-8); 131.62 (C-5); 73.94 (C-1″); 66.37 (C-3′); 29.43 (d, $J_{4'-P}$ = 129.7 Hz, C-4′). For $C_8H_{10}ClN_4O_4P$ (292.62) calcd: C, 32.84; H, 3.44; N, 19.15. Found: C, 32.75; H, 3.71; N, 19.06.

4.1.9.73. 2-Amino-6-chloro-9-[1-(phosphonoethoxy)methyl]-9*H*-purine (17b). Method A: compound **14b**, 0.5 g (1.3 mmol); HPLC; obtained 0.17 g (43%) of compound **17b**. ESI [M−H] 305.9 (100).^{1}H NMR (D_2O): 8.25 (s, 1H, H-8); 5.56 (s, 2H, H-1′); 3.78 (m, 2H, H-3′); 1.84 (m, 2H, H-4′). ^{13}C NMR (D_2O): 160.33 (C-2); 154.07 (C-4); 151.24 (C-6); 145.06 (C-8); 124.43 (C-5); 73.17 (C-1′); 66.98 (C-3′); 30.12 (d, $J_{4'-P}$ = 126.1 Hz, C-4′). For $C_8H_{11}ClN_5O_4P$. 1/5 H_2O (307.63) calcd: C, 30.87; H, 3.69; N, 22.50. Found: C, 30.89; H, 3.70; N, 22.62.

4.1.9.74. 6-Chloro-9-[1-(phosphonopropoxy)methyl]-9*H*-purine (17c). Method A: compound **14c**, 0.5 g (1.3 mmol); HPLC; obtained 0.22 g (56%) of compound **17c**. ESI [M+H] 306.8 (50).^{1}H NMR (D_2O): 8.77 (s, 1H, H-2); 8.71 (s, 1H, H-8); 5.79 (s, 2H, H-1′); 3.62 (t, 2H, $J_{3'-4'}$ = 6.7 Hz, H-3′); 1.75 (m, 2H, H-4′); 1.33 (m, 2H, H-5′). ^{13}C NMR (D_2O): 152.59 (C-2); 152.21 (C-4); 150.86 (C-6); 148.38 (C-8); 131.60 (C-5); 73.99 (C-1′); 71.79 (d, $J_{3'-P}$ = 19.1 Hz, C-3′); 26.06 (d, $J_{5'-P}$ = 131.5 Hz, C-5′); 24.71 (d, $J_{4'-P}$ = 3.8 Hz, C-4′). For $C_9H_{12}ClN_4O_4P$ (306.64) calcd: C, 35.25; H, 3.94; N, 18.27. Found: C, 35.04; H, 4.05; N, 17.93.

4.1.9.75. 2-Amino-6-chloro-9-[1-(phosphonopropoxy)methyl]-9*H*-purine (17d). Method A: compound **14d**, 0.5 g (1.2 mmol); HPLC; obtained 0.24 g (61%) of compound **17d**. ESI [M−H] 320.3 (100).^{1}H NMR (D_2O): 8.21 (s, 1H, H-8); 5.54 (s, 2H, H-1′); 3.60 (t, 2H, $J_{3'-4'}$ = 6.5, H-3′); 1.76 (m, 2H, H-4′); 1.48 (m, 2H, H-5′). ^{13}C NMR (D_2O): 160.24 (C-2); 153.95 (C-4); 151.15 (C-6); 144.92 (C-8); 124.21 (C-5); 73.30 (C-1′); 70.78 (d, $J_{3'-P}$ = 19.0 Hz, C-3′); 25.23 (d, $J_{5'-P}$ = 133.6 Hz, C-5′); 24.03 (d, $J_{4'-P}$ = 3.7 Hz, C-4′). For $C_9H^{13}ClN_5O_4P$. 1/3 H_2O (321.66) calcd: C, 32.99; H, 4.20; N, 21.37. Found: C, 33.03; H, 4.19; N, 21.13.

4.1.9.76. 6-chloro-9-[1-(phosphonobutoxy)methyl]-9*H*-purine (17e). Method A: compound **14e**, 1.35 g (3.3 mmol); HPLC; obtained 0.4 g (37%) of compound **17e**. ESI [M−H] 318.9 (100). ^{1}H

NMR (D_2O+NaOD): 8.77 (s, 1H, H-8); 8.71 (s, 1H, H-2); 5.79 (s, 2H, H-1′); 3.63 (t, 2H, $J_{3'-4'}$ = 6.7 Hz, H-3′); 1.60 (p, 2H, $J_{4'-3'}$ = $J_{4'-5'}$ = 7.1 Hz H-4′); 1.48 (m, 2H, H-5′); 1.34 (m, 2H, H-6′). ^{13}C NMR (D_2O+NaOD): 152.65 (C-2); 152.21 (C-4); 150.87 (C-6); 148.37 (C-8); 131.56 (C-5); 74.05 (C-1′); 70.59 (C-3′); 30.92 (d, $J_{4'-P}$ = 17.0 Hz, C-4′); 29.63 (d, $J_{6'-P}$ = 130.6 Hz, C-6′); 21.22 (d, $J_{5'-P}$ = 3.8 Hz, C-5′). For $C_{10}H_{14}ClN_4O_4P$ (320.67) calcd: C, 37.46; H, 4.40; N, 17.47. Found: C, 37.43; H, 4.61; N, 17.18.

4.1.9.77. 2-Amino-6-chloro-9-[1-(phosphonobutoxy)methyl]-9*H*-purine (17f). Method A: compound **14f**, 1.85 g (4.4 mmol); HPLC; obtained 0.59 g (40%) of compound **17f**. ESI [M−H] 334.0 (100).^{1}H NMR (D_2O+NaOD): 8.24 (s, 1H, H-8); 5.56 (s, 2H, H-1′); 3.60 (t, 2H, $J_{3'-4'}$ = 6.7 Hz, H-3′); 1.59 (m, 2H, H-4′); 1.47 (m, 2H, H-5′); 1.35 (m, 2H, H-6′). ^{13}C NMR (D_2O+NaOD): 160.37 (C-2); 154.13 (C-4); 151.25 (C-6); 145.07 (C-8); 124.39 (C-5); 73.35 (C-1′); 70.25 (C-3′); 30.93 (d, $J_{4'-P}$ = 14.0 Hz, C-4′); 29.65 (d, $J_{6'-P}$ = 130.9 Hz, C-6′); 21.24 (d, $J_{5'-P}$ = 3.8 Hz, C-5′). For $C_{10}H_{15}ClN_5O_4P$. ¼ H_2O (316.25) calcd: C, 35.31; H, 4.59; N, 20.59. Found: C, 35.57; H, 4.64; N, 20.23.

4.1.9.78. 9-[1-(Phosphonoethoxy)methyl]xanthine (18a). Method E: Compound **16b**, 0.40 g (1.4 mmol); HPLC, crystallized from H_2O obtained 0.23 g (57%) of compound**18a**. ESI [M−H] 288.9 (100).^{1}H NMR (D_2O): 7.86 (s, 1H, H-8); 5.47 (s, 2H, H-1′); 3.77 (dm, 2H, $J_{3'-P}$ = 10.2 Hz, H-3′); 1.92 (dm, 2H, $J_{4'-P}$ = 18.0 Hz, H-4′). ^{13}C NMR (D_2O): 161.32 (C-6); 158.00 (C-2); 150.08 (C-4); 139.73 (C-8); 115.76 (C-5); 73.43 (C-1′); 65.41 (C-3′); 29.22 (d, $J_{4'-P}$ = 130.4 Hz, C-4′). For $C_8H_{11}N_4O_6P$. 1/2 H_2O (290.17) calcd: C, 32.12; H, 4.04; N, 18.73. Found: C, 32.07; H, 3.98; N, 18.63.

4.1.9.79. 9-[1-(Phosphonopropoxy)methyl]xanthine (18b). Method E: Compound **16d,** 0.25 g (0.8 mmol); HPLC, crystallized from H_2O obtained 0.12 g (48%) of compound **18b**. ESI [M−H] 302.9 (100).^{1}H NMR (D_2O): 7.85 (s, 1H, H-8);5.46 (s, 2H, H-1′); 3.55 (m, 2H, H-3′); 1.70 (m, 2H, H-4′); 1.35 (m, 2H, H-5′).13C NMR (D_2O): 161.50 (C-6);160.16 (C-2); 152.17 (C-4); 139.75 (C-8); 115.47 (C-5);72.90 (C-1′); 71.00 (d, $J_{3'-P}$ = 19.4 Hz, C-3′); 26.14 (d, $J_{5'-P}$ = 131.4 Hz, C-5′); 24.70 (d, $J_{4'-P}$ = 3.5 Hz, C-4′). For $C_9H_{13}N_4O_6P$; 2/5 H_2O (304.20) calcd: C, 32.46; H, 4.90; N, 16.82. Found: C, 32.13; H, 4.65; N, 16.81.

4.1.9.80. 9-[1-(Phosphonobutoxy)methyl]xanthine (18c). Method E: Compound **16f** 0.12 g (0.4 mmol); HPLC, crystallized from H_2O obtained 0.07 g (58%) of compound **18c**. ESI [M+Na] 340.9 (100).^{1}H NMR (D_2O): 7.83 (s, 1H, H-8); 5.43 (s, 2H, H-1′); 3.59 (t, 2H, $J_{3'-4'}$ = 6.7 Hz, H-3′); 1.60 (m, 2H, H-4′); 1.48 (m, 2H, H-5′); 1.35 (m, 2H, H-6′). ^{13}C NMR (D_2O):161.91 (C-6); 160.77 (C-2); 154.40 (C-4); 139.86 (C-8); 115.36 (C-5); 72.85 (C-1′); 69.87 (C-3′); 30.99 (d, $J_{4'-P}$ = 17.0 Hz, C-4′); 29.70 (d, $J_{6'-P}$ = 130.7 Hz, C-6′); 21.27 (d, $J_{5'-P}$ = 3.8 Hz, C-5′). For $C_{10}H_{15}N_4O_6P$ (318.22) calcd: C, 33.90; H, 5.41; N, 15.82. Found: C, 33.56; H, 5.65; N, 15.78.

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Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bmc.2011.11.034.

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