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Tricyclic etheno analogs of PMEG and PMEDAP: Synthesis and biological activity

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Abstract—A series of novel 9-substituted (2-(3*H*-imidazo[1,2-*a*]purin-3-yl)ethoxy)methylphosphonic and 4-substituted (2-(1*H*-imidazo[2,1-*b*]purin-1-yl)ethoxy)methylphosphonic acids as tricyclic etheno analogs of potent antivirals and cytostatics PMEG and PMEDAP was synthesized and evaluated for their biological activity. Most of the compounds showed modest activity against varicella-zoster virus (VZV) and human cytomegalovirus (HCMV) except for (2-(9-oxo-5,9-dihydro-3*H*-imidazo[1,2-*a*]purin-3-yl)ethoxy)methylphosphonic acid **8** which proved markedly active against VZV and HCMV. None of the compounds tested exhibited any significant cytostatic effect.

1. Introduction

Acyclic nucleoside phosphonates (ANPs) represent a key class of nucleotide analogs with broad spectrum of biological activity, either antiviral, antineoplastic, or antiprotozoal.[1]

The highlights of the ANP series are EMEA and FDA-approved antiviral drugs PMEA (adefovir, formulated as adefovir dipivoxil, Hepsera™), (S)-HPMPC (cidofovir, Vistide™), and (*R*)-PMPA (tenofovir, formulated as tenofovir disoproxil fumarate, Viread™).[2] Another important member of the family of acyclic nucleoside phosphonates, PMEDAP (**1**, Fig. 1), is a broad-spectrum antiviral agent with potent activity against DNA viruses. Despite its excellent antiviral parameters, it was never systemically pursued in preclinical phase of drug development, mainly due to its cytotoxicity. Its main importance now lies in the antitumor therapy. Similarly, PMEG (**2**) is an extremely active antiviral, but again handicapped for clinical use by its toxicity.[3,4] Both PMEDAP (**1**) and PMEG (**2**) are phosphorylated by cellular nucleotide kinases. The corresponding diphosphates (PMEDAPpp and PMEGpp) inhibit cellular DNA polymerases and its incorporation in the growing DNA chain leads to chain termination.[5–7] In addition, PMEG and its anabolites efficiently inhibit purine nucleoside phosphorylases;[8] this activity probably contributes to their high toxicity. PMEDAP (**1**) as well as PMEG (**2**) exhibit potent cytostatic activity in vivo in rat and mouse carcinomas and sarcomas.[9–11] Some of the N^6-substituted PMEDAP derivatives show marked cytostatic activity in vivo or in vitro.[12,13]

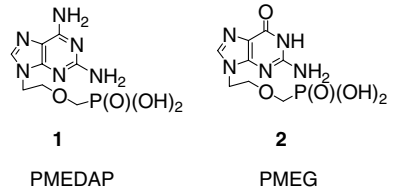

Figure 1. PMEDAP and PMEG structures.

In this paper, we describe the synthesis of tricyclic etheno analogs of PMEDAP and PMEG, and their antiviral and cytostatic properties. The tricyclic etheno-bridged purine derivatives result from the reaction of purines bearing at least one amino group on the pyrimidine ring with chloroacetaldehyde or its synthetic congeners.[14–16]

Keywords: Acyclic nucleoside phosphonates; Antiviral; Chloroacetaldehyde; Purines.
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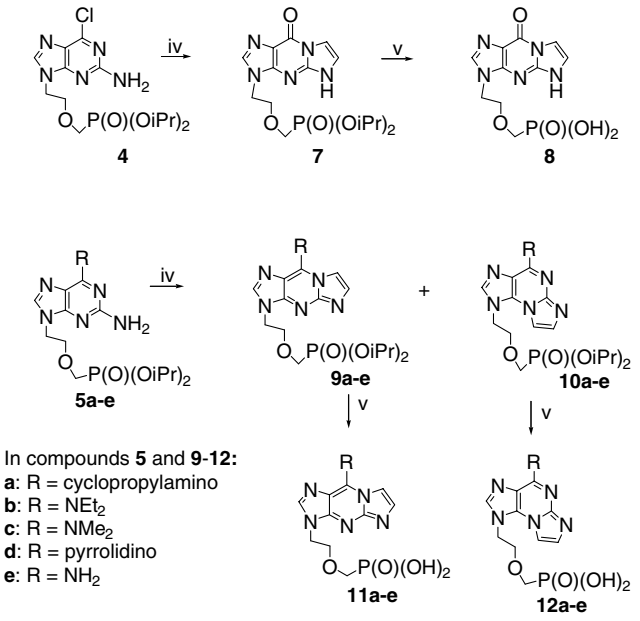
Generally, two sites of cyclization are possible, giving rise to linear and angular isomer, respectively.[14,15,17] Among the etheno-bridged purines, the most intensely studied are the 1,N^6-ethenoadenine derivatives,[18–25] while there are relatively few scientific papers concerning ethenopurines derived from 2,6-diaminopurine or 2-aminopurine.[17,26,27,32] Some time ago, an analogous study of biological activity of tricyclic etheno derivatives of acyclovir and ganciclovir was performed. [28,29] These tricyclic derivatives were less potent antivirals, but they showed marked selectivity toward some of the DNA viruses, namely HSV-1 and HSV-2.[30] In the view of these results, we were interested to find out whether the introduction of the etheno bridge to PMEDAP (**1**) and PMEG (**2**) would lead to modulation of their activity, selectivity or cytotoxicity.

2. Chemistry

There are different synthetic approaches toward the etheno-bridged purine analogs based on the reaction of aminopurines with aqueous chloroacetaldehyde,[14–16] anhydrous haloacetaldehydes,[31] or bromoacetaldehyde diethyl acetal.[31] For the synthesis of our etheno-bridged PMEDAP and PMEG analogs, we have decided to use the method employing aqueous chloroacetaldehyde that we recently optimized for the cyclization of 2-amino-9-methylpurines affording the etheno-bridged products in reasonable yields.[32]

In the present work, the cyclizations were performed with diisopropyl esters of 2-amino-9-(phosphonomethoxyethyl)purines **4** and **5a**–**e**. The starting compounds **4** and **5e** were prepared by alkylation of 2-amino-6-chloropurine **3** and 2,6-diaminopurine **6** with the phosphonomethoxyethyl synthon (Scheme 1).[3] Further nucleophilic substitution of chloro intermediate **4** by various amines led to series of N^6-substituted PMEDAP derivatives **5a**–**d**.[4]

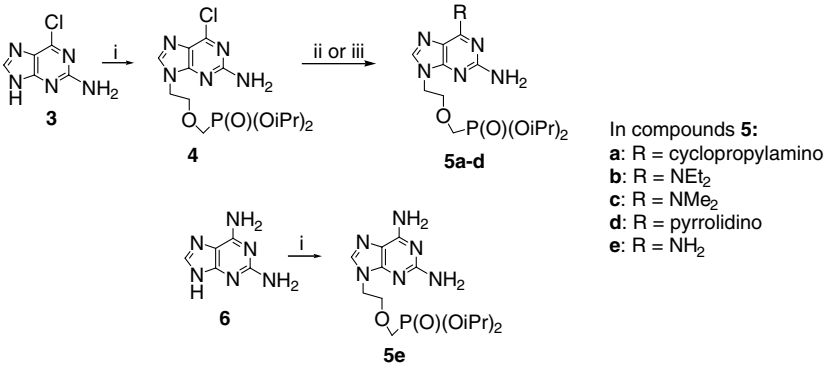
First, we have examined the cyclization of 2-amino-6-chloropurine derivative **4** with chloroacetaldehyde (Scheme 2). The reaction was carried out at pH 6 and 70 °C, in a mixture of water–dioxane as solvents, under the same conditions as those we have described previously for 2-amino-9-methylpurines.[32] Apart from the cyclization, hydrolysis of C–Cl bond took place leading directly and exclusively to linear etheno-bridged PMEG analogue **7** in a good yield (Table 1, entry 1). The exclusive formation of linear regioisomer is explained by the stabilization in the form of 5-*H* tautomer and corresponds to our previous results from the 2-amino-9-methylpurine series.[32] The target tricyclic PMEG deriv-



Scheme 2. Synthesis of etheno-bridged PMEDAP and PMEG derivatives. Reagents and conditions: (iv) 1 M aq $ClCH_2CHO$, water/dioxane, 70 °C, 8 h, for yields, see Table 1; (v) Me_3SiBr, acetonitrile, rt, 24 h, for yields, see Section 5.

Table 1. Yields of linear and angular isomers

Entry	Starting material	Linear product	Yield of linear isomer (%)	Angular product	Yield of angular isomer (%)
1	**4**	**7**	54	—	0
2	**5a**	**9a**	36	**10a**	0
3	**5b**	**9b**	0	**10b**	48
4	**5c**	**9c**	28	**10c**	49
5	**5d**	**9d**	22	**10d**	36
6	**5e**	**9e**	15	**10e**	43



Scheme 1. Synthesis of starting compounds. Reagents and conditions: (i) 1—0.5 equiv Cs_2CO_3, DMF; 2—1.2 equiv $Cl(CH_2)_2OCH_2P(O)(O\text{-}i\text{-}Pr)_2$, 80 °C, 8 h, 50–56%; (ii) 3 equiv $(CH_3)_2NH(CH_3)_2NCOOH$, acetonitrile, reflux, 3 h, 75%; (iii) 4 equiv amine RH, abs EtOH, reflux, 3 h, 65–80%.

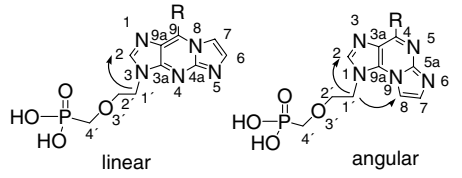


Figure 2. General numbering and NOE contacts for the linear and angular isomers.

ative **8** was obtained after cleavage of isopropyl esters of intermediate **7** by Lewis acid (Scheme 2).

Cyclizations of PMEDAP derivatives **5a**–**e** with chloroacetaldehyde were further studied (Scheme 2). Regioselectivity of the reaction was strongly dependent on the character of the amino substituent in position 6 of purine scaffold. Cyclization of 6-cyclopropylaminopurine derivative **5a** provided exclusively the linear tricyclic system **9a** (Table 1, entry 2), again probably due to the stabilization in the form of 5-*H* tautomer. However, in the case of cyclization of 6-diethylaminopurine **5b** the angular product **10b** was the only product observed (entry 3). In all the other cases, 6-dimethylamino **5c**, 6-pyrrolidino **5d**, and 6-aminopurine **5e**, both linear **9** and angular **10** products were isolated with some predominance of the angular one (entries 4–6).

The overall yields of the cyclizations were good to very good (Table 1) and higher than those of previously reported similar cyclizations[14,30,31] and even higher than those of corresponding 9-methylpurine derivatives.[32] Target-free tricyclic PMEDAP derivatives **11** and **12** were again obtained after cleavage of isopropyl esters by Lewis acid (Scheme 2).

For structure elucidation of the new compounds, NMR spectroscopy was used. While the complete assignment of all ^{1}H and ^{13}C signals was based on a combination of ^{1}H, ^{13}C APT, H,C-HSQC, and H,C-HMBC experiments, a DPFGSE NOE experiment was found to be the most efficient way to distinguish between the linear and angular isomers (Fig. 2).[32]

3. Biological activity

3.1. Antiviral activity

All the free nucleotide analogs **8**, **11a**, **11c**, **11d**, **11e**, **12b**, **12c**, **12d**, and **12e** were examined for their inhibitory effect on the replication of herpes viruses, namely varicella-zoster virus (VZV) and human cytomegalovirus (HCMV), in human embryonic lung (HEL) cells (Table 2). One of the parent compounds, PMEDAP (**1**), is highly active against HSV-1 and HSV-2 within the 50% effective concentration (EC_{50}) range of 0.00024–0.007 μmol/mL. Against VZV and CMV it exhibited inhibitory activity within the EC_{50} range of 0.0035–0.035 μmol/mL.[3] The other parent compound, PMEG (**2**), exhibits potent activity against all mentioned DNA viruses ($EC_{50} \approx 0.000034$–0.00007 μmol/mL), but it is significantly toxic against the host cells.[3]

Table 2. Antiviral activity-EC_{50} (μmol/ml) and cytotoxicity (μmol/ml)

Compound	Antiviral activity-EC_{50} (μmol/ml)[a]						Cytotoxicity (μmol/ml)	
	VZV				HCMV		Cell morphology[b] (MCC)	Cell growth[c] (CC_{50})
	TK^+ strain		TK^- strain		AD-169 strain	Davis strain		
	YS strain	Oka strain	07/1 strain	YS/R strain				
8	0.0016	0.00073 ± 0	0.0020 ± 0.0008	0.000084	0.013 ± 0.006	0.0039 ± 0.0017	>0.28	0.12
11a	0.0077	0.0062 ± 0.001	0.0088 ± 0.004	0.003	0.026 ± 0.016	0.01 ± 0.001	⩾0.28	0.14
11c	0.011	0.0035 ± 0.003	0.0073 ± 0.005	0.0038	0.042 ± 0.024	0.013 ± 0.002	>0.29	>0.15
11d	0.017	0.0038 ± 0.001	0.011 ± 0.003	0.0014	0.032 ± 0.013	0.014 ± 0.011	>0.27	>0.14
11e	ND[d]	0.064	0.147	ND	>0.32	0.186	>0.32	>0.16
12b	ND	>0.27	>0.27	ND	>0.27	>0.27	>0.27	>0.14
12c	0.026	0.018 ± 0.006	0.027 ± 0.003	0.018	0.098 ± 0.027	0.034 ± 0.01	>0.29	>0.15
12d	0.018	0.011 ± 0.003	0.02 ± 0.009	0.0076	0.033 ± 0.011	0.018 ± 0.006	>0.27	>0.14
12e	0.051	0.031 ± 0.024	0.053 ± 0.042	0.025	0.144	0.118	>0.32	>0.16
Acyclovir	0.0043 ± 0.00009	0.0016 ± 0.0008	0.082 ± 0.009	0.06 ± 0.009	ND	ND	>0.22	>0.71
Ganciclovir	ND	ND	ND	ND	0.013 ± 0.012	0.0074 ± 0.007	⩾1.57	0.47 ± 0.3
Cidofovir	ND	ND	ND	ND	0.0013 ± 0.0007	0.00075 ± 0.0007	⩾1.43	0.23 ± 0.12

[a] Effective concentration required to reduce virus plaque formation by 50%. Virus input was 20 PFU (VZV) or 100 PFU (HCMV).
[b] Minimum cytotoxic concentration that causes a microscopically detectable alteration of cell morphology.
[c] Cytotoxic concentration required to reduce cell growth by 50%.
[d] Not determined.

Compounds **12b** and **11e** proved to be inactive against both VZV and HCMV. Compounds **11a**, **11c**, **11d**, **12c**, **12d**, and **12e** exhibited activity against both wild-type and thymidine kinase-deficient (TK^-) strain of VZV with EC_{50} values ranging from 0.003 to 0.053 μmol/mL. These compounds exhibited lower activity against CMV, since EC_{50} values ranged from 0.004 to 0.144 μmol/mL. The best results were obtained for the PMEG analog **8** which exhibited significant activity against both VZV and CMV (see Table 2). Thus, EC_{50} values for analog **8** against TK^+ strains of VZV were similar to those obtained for the reference compound acyclovir. In contrast to acyclovir, compound **8** proved equally active against TK^+ and TK^- VZV strains. Also, the PMEG analog **8** showed anti-HCMV activity comparable to that observed for the reference compound ganciclovir. Furthermore, all the compounds were subjected to cytotoxicity measurements based on the following parameters: alteration of cell morphology and inhibition of cell growth (see Table 2). All of the test compounds did not affect cell morphology up to a concentration of 0.27 μmol/mL and CC_{50} values for cell growth were $\geqslant$0.12 μmol/mL.

3.2. Cytostatic activity

Compounds **8**, **11a**, **11c**, **11d**, **11e**, **12b**, **12c**, **12d**, and **12e** were tested for cytostatic effect on the cultures of murine leukemia L1210 cells, murine L929 cells, human cervix carcinoma HeLa S3 cells, and human T-lymphoblastoid CCRF-CEM cell line. None of the samples exhibited any significant activity in these cell culture assays.

4. Conclusion

We have synthesized a series of novel tricyclic etheno analogs of PMEDAP and PMEG by cyclization of derivatives of parent compounds with chloroacetaldehyde. Most of target compounds showed only modest activity against DNA viruses, with the exception of **8**, a tricyclic PMEG analog, which exhibited a pronounced inhibitory activity against VZV and HCMV. None of the compounds possessed any significant cytostatic activity.

5. Experimental

5.1. General

Melting points were determined on a Kofler block and are uncorrected. Analytical TLC was performed on silica gel pre-coated aluminum plates with fluorescent indicator (Merck 5554, 60 F_{254}). Spots were visualized with UV light (254 nm). Column chromatography was carried out on silica gel (Sigma S-0507, 40–63 μm). Mass spectra were measured on a ZAB-EQ (Micromass, Manchester, UK) spectrometer using the FAB (ionization with Xe, accelerating voltage 8 kV, thioglycerol/glycerol, 3:1 mixture or bis(2-hydroxyethyl) disulfide was used as matrix). ^1H and ^{13}C NMR spectra were recorded at 500 and 125.7 MHz on a Varian Unity 500 instrument in DMSO-d_6 (referenced to the solvent signal δ = 2.50 and 39.70 ppm, respectively). Chemical shifts are in ppm and coupling constants (J) in Hz. UV spectra were taken on a Beckman DU-65 spectrophotometer in methanol or water solution. IR spectra were obtained on an FT IR Bruker Equinox IFS 55 spectrometer in KBr pellets.

5.2. Alkylation of 2-amino-6-chloropurine and 2,6-diaminopurine

Compounds **4** and **5e** were prepared by alkylation of 2-amino-6-chloropurine (**3**) or 2,6-diaminopurine (**6**), respectively, with diisopropyl[(2-chloroethoxy)methyl]phosphonate as described in Ref. 3.

5.3. Preparation of N^6-substituted 2,6-diamino-9-[2-(phosphonomethoxy)ethyl]purines

Compounds **5a**–**d** were prepared from **4** by heating with the corresponding primary or secondary amine in dry ethanol or, in case of dimethylamino derivative **7c**, by reflux with dimethylammonium *N*,*N*-dimethylcarbamate in dry acetonitrile, as described in Ref. 4. Products were purified by flash chromatography and used directly in the cyclization reaction with chloroacetaldehyde. The following compounds were prepared by this procedure:

5.4. Diisopropyl (2-(2-amino-6-(cyclopropylamino)-9*H*-purin-9-yl)ethoxy)methylphosphonate (5a)

Prepared from **4** (780 mg, 2 mmol) and cyclopropylamine (450 mg, 8 mmol, 4 equiv). Yield: 530 mg (65%) of **5a** as brown syrupy product. MS (FAB) *m*/*z* (rel intensity): 413 (100, M+1). HR MS (FAB): for $C_{17}H_{29}N_6O_4P$ calcd 413.2066, found 413.2050. ^1H NMR (400 MHz, DMSO-d_6): 0.54–0.68 (m, 4H, CH_2-cycloprop); 1.15 and 1.19 (2× d, 2× 6H, J_{vic} = 6.2, $(CH_3)_2CH$); 3.02 (br m, 1H, CH–N); 3.76 (d, 2H, $J_{H,P}$ = 8.4, H-4′); 3.82 (t, 2H, $J_{2',1'}$ = 5.2, H-2′); 4.13 (t, 2H, $J_{1',2'}$ = 5.2, H-1′); 4.51 (dh, 2H, $J_{H,P}$ = 7.7, J_{vic} = 6.2, $CH(CH_3)_2$); 5.82 (br s, 2H, NH_2); 7.27 (br s, 1H, NH); 7.64 (s, 1H, H-8). ^{13}C NMR (100.6 MHz, DMSO-d_6): 6.61 (CH_2-cycloprop); 23.83 and 23.96 (d, $J_{C,P}$ = 4, $(CH_3)_2CH$); 42.06 (CH_2-1′); 64.78 (d, $J_{C,P}$ = 164, CH_2-4′); 70.38 (d, $J_{C,P}$ = 6, $CH(CH_3)_2$); 70.59 (d, $J_{C,P}$ = 12, CH_2-2′); 113.47 (C-5); 137.59 (CH-8); 151.43 (C-4); 156.06 (C-6); 160.32 (C-2).

5.5. Diisopropyl (2-(2-amino-6-(diethylamino)-9*H*-purin-9-yl)ethoxy)methylphosphonate (5b)

Prepared from **4** (780 mg, 2 mmol) and diethylamine (580 mg, 8 mmol, 4 equiv). Yield: 685 mg (80%) of **5b** as white crystalline product. MS (FAB) *m*/*z* (rel intensity): 429 (100, M+1). ^1H NMR (400 MHz, DMSO-d_6, 80 °C): 1.17 (t, 6H, J_{vic} = 7.0, CH_3CH_2); 1.18 and 1.21 (2× d, 2× 6H, J_{vic} = 6.2, $(CH_3)_2CH$); 3.74 (d, 2H, $J_{H,P}$ = 8.2, H-4′); 3.85 (t, 2H, $J_{2',1'}$ = 5.3, H-2′); 3.88 (q, 4H, J_{vic} = 7.0, CH_2CH_3); 4.14 (t, 2H, $J_{1',2'}$ = 5.3, H-1′); 4.54 (dh, 2H, $J_{H,P}$ = 7.7, J_{vic} = 6.2, $CH(CH_3)_2$); 5.43 (br s, 2H, NH_2); 7.64 (s, 1H, H-8). ^{13}C NMR (100.6 MHz, DMSO-d_6, 80 °C): 13.45 (CH_3CH_2); 23.50 and 23.61 (d, $J_{C,P}$ = 4, $(CH_3)_2CH$); 41.65 (CH_2CH_3);

41.95 (CH_2-1′); 64.98 (d, $J_{C,P}$ = 165, CH_2-4′); 70.28 (d, $J_{C,P}$ = 6, $CH(CH_3)_2$); 70.34 (d, $J_{C,P}$ = 11, CH_2-2′); 114.64 (C-5); 136.78 (CH-8); 152.67 (C-4); 153.69 (C-6); 159.47 (C-2). For $C_{18}H_{33}O_4P$ calcd 50.46% C, 7.76% H, 19.61% N, 7.23% P, found 50.26% C, 7.74% H, 19.38% N, 7.01% P.

5.6. Diisopropyl (2-(2-amino-6-(dimethylamino)-9*H*-purin-9-yl)ethoxy)methylphosphonate (5c)

Prepared from **4** (780 mg, 2 mmol) and *N*,*N*-dimethylammoniumdimethylcarbamate (0.8 mL, 6 mmol, 3 equiv). Yield: 620 mg (75%) of **5c** as yellow solid. MS (FAB) *m*/*z* (rel intensity): 401 (100, M+1). ^{1}H NMR (400 MHz, DMSO-d_6): 1.16 and 1.19 (2× d, 2× 6H, J_{vic} = 6.2, $(CH_3)_2CH$); 3.36 (br s, 6H, $(CH_3)_2N$); 3.76 (d, 2H, $J_{H,P}$ = 8.4, H-4′); 3.82 (t, 2H, $J_{2',1'}$ = 5.3, H-2′); 4.14 (t, 2H, $J_{1',2'}$ = 5.3, H-1′); 4.51 (dh, 2H, $J_{H,P}$ = 7.7, J_{vic} = 6.2, $CH(CH_3)_2$); 5.81 (br s, 2H, NH_2); 7.66 (s, 1H, H-8). ^{13}C NMR (100.6 MHz, DMSO-d_6): 23.81 and 23.94 (d, $J_{C,P}$ = 4, $(CH_3)_2CH$); 37.77 ($(CH_3)_2N$); 42.13 (CH_2-1′); 64.82 (d, $J_{C,P}$ = 164, CH_2-4′); 70.41 (d, $J_{C,P}$ = 6, $CH(CH_3)_2$); 70.51 (d, $J_{C,P}$ = 11, CH_2-2′); 113.75 (C-5); 136.93 (CH-8); 152.84 (C-4); 154.94 (C-6); 159.64 (C-2). For $C_{16}H_{29}N_6O_4P$ calcd 47.99% C, 7.30% H, 20.99% N, 7.74% P, found 47.68% C, 7.25% H, 20.72% N, 7.40% P.

5.7. Diisopropyl (2-(2-amino-6-(pyrrolidin-1-yl)-9*H*-purin-9-yl)ethoxy)methylphosphonate (5d)

Prepared from **4** (780 mg, 2 mmol) and pyrrolidine (570 mg, 8 mmol, 4 equiv). Yield: 600 mg (71%) of **5d** as brown syrupy product. MS (FAB) *m*/*z* (rel intensity): 427 (100, M+1). HR MS (FAB): for $C_{18}H_{31}N_6O_4P$ calcd 427.2222, found 427.2232. ^{1}H NMR (400 MHz, DMSO-d_6): 1.16 and 1.20 (2× d, 2× 6H, J_{vic} = 6.2, $(CH_3)_2CH$); 1.89 (br m, 4H, CH_2-pyrrolidine); 3.60 (br m, 2H, CH_2N-pyrrolidine); 3.75 (d, 2H, $J_{H,P}$ = 8.4, H-4′); 3.81 (t, 2H, $J_{2',1'}$ = 5.3, H-2′); 3.90 (br m, 2H, CH_2N-pyrrolidine); 4.13 (t, 2H, $J_{1',2'}$ = 5.3, H-1′); 4.52 (dh, 2H, $J_{H,P}$ = 7.7, J_{vic} = 6.2, $CH(CH_3)_2$); 5.77 (br s, 2H, NH_2); 7.63 (s, 1H, H-8). ^{13}C NMR (100.6 MHz, DMSO-d_6): 23.81 and 23.94 (d, $J_{C,P}$ = 4, $(CH_3)_2CH$); 24.00 and 25.40 (CH_2-pyrrolidine); 42.03 (CH_2-1′); 47.40 (CH_2N-pyrrolidine); 64.81 (d, $J_{C,P}$ = 164, CH_2-4′);70.36 (d, $J_{C,P}$ = 6, $CH(CH_3)_2$); 70.53 (d, $J_{C,P}$ = 12, CH_2-2′); 113.92 (C-5); 137.30 (CH-8); 152.39 (C-4); 153.24 (C-6); 159.97 (C-2).

5.8. Cyclization reaction with chloroacetaldehyde. General procedure

Compounds **7**, **9a**–**e** and **10a**–**e** were prepared by cyclization of compounds **4**, **5a**–**e** with 1 M aq solution of 2-chloroacetaldehyde in mixture of water and dioxane under heating to 70 °C for 7 h.[32]

5.9. Diisopropyl (2-(9-oxo-5,9-dihydro-3*H*-imidazo[1,2-*a*]purin-3-yl)ethoxy)methylphosphonate (7)

Prepared from **4** (220 mg, 0.56 mmol) and chloroacetaldehyde (15 ml, 1 M aq solution). Chromatography over silica gel in MeOH/$CHCl_3$ 3:97 yielded 120 mg (54%) of **7** as yellow syrupy product. MS (FAB) *m*/*z* (rel intensity): 398 (100, M+1). HR MS (FAB): for $C_{16}H_{24}N_5O_5P$ calcd 398.1593, found 398.1605. UV (MeOH): 281 (7960), 224 (22500). FT IR (KBr): 3423, 3130, 2980, 2932, 2854, 2749, 1698, 1605, 1562, 1549, 1529, 1456, 1386, 1376, 1252, 1238, 1200, 1106, 991, 889. ^{1}H NMR (400 MHz, DMSO-d_6): 1.11 and 1.15 (2× d, 2× 6H, J_{vic} = 6.2, $(CH_3)_2CH$); 3.78 (d, 2H, $J_{H,P}$ = 8.4, H-4′); 3.89 (t, 2H, $J_{2',1'}$ = 5.4, H-2′); 4.26 (t, 2H, $J_{1',2'}$ = 5.4, H-1′); 4.48 (dh, 2H, $J_{H,P}$ = 7.7, J_{vic} = 6.2, $CH(CH_3)_2$); 7.44 (d, 1H, $J_{6,7}$ = 2.6, H-6); 7.61 (d, 1H, $J_{7,6}$ = 2.6, H-7); 7.93 (s, 1H, H-2); 12.48 (br s, 1H, NH). ^{13}C NMR (100.6 MHz, DMSO-d_6): 23.75 and 23.90 (d, $J_{C,P}$ = 4, $(CH_3)_2CH$); 42.73 (CH_2-1′); 64.74 (d, $J_{C,P}$ = 164, CH_2-4′); 70.34 (d, $J_{C,P}$ = 6, $CH(CH_3)_2$); 71.15 (d, $J_{C,P}$ = 11, CH_2-2′); 107.00 (CH-7); 114.73 (C-9a); 116.61 (CH-6); 139.49 (CH-2); 146.12 (C-4a); 150.48 (C-3a); 151.34 (C-9).

5.10. Diisopropyl (2-(9-(cyclopropylimino)-5,9-dihydro-3*H*-imidazo[1,2-*a*]purin-3-yl)ethoxy)methylphosphonate (9a)

Prepared from **5a** (0.9 g, 2.2 mmol) and chloroacetaldehyde (20 ml, 1 M aq solution). Chromatography over silica gel in MeOH/$CHCl_3$ 2:98 afforded 0.34 g (36%) of **9a** as brown syrupy product. MS (FAB) *m*/*z* (rel intensity): 437 (100, M+1). HR MS (FAB): for $C_{19}H_{29}N_6O_4P$ calcd 437.2066, found 437.2076. UV (MeOH): 328 (5780), 277 (4170), 239 (24630). FT IR (KBr): 3430, 3077, 2980, 2934, 2749, 1662, 1584, 1559, 1529, 1440, 1386, 1376, 1362, 1240, 1177, 1142, 1106, 990, 889. ^{1}H NMR (500 MHz, DMSO-d_6): 0.94 and 1.03 (2× m, 2× 2H, CH_2-cycloprop); 1.12 and 1.16 (2× d, 2× 6H, J_{vic} = 6.2, $(CH_3)_2CH$); 3.80 (d, 2H, $J_{H,P}$ = 8.3, H-4′); 3.85 (tt, 1H, J = 7.1, 3.9, CH–N); 3.94 (t, 2H, $J_{2',1'}$ = 5.2, H-2′); 4.38 (t, 2H, $J_{1',2'}$ = 5.2, H-1′); 4.49 (dh, 2H, $J_{H,P}$ = 7.7, J_{vic} = 6.2, $CH(CH_3)_2$); 7.87 (d, 1H, $J_{6,7}$ = 2.7, H-6); 8.32 (s, 1H, H-2); 8.80 (d, 1H, $J_{7,6}$ = 2.7, H-7); 10.00 (br s, 1H, NH). ^{13}C NMR (125.8 MHz, DMSO-d_6): 7.48 (CH_2-cycloprop); 23.75 and 23.88 (d, $J_{C,P}$ = 4, $(CH_3)_2CH$); 27.95 (CH–N); 42.92 (CH_2-1′); 64.72 (d, $J_{C,P}$ = 164, CH_2-4′); 69.94 (d, $J_{C,P}$ = 12, CH_2-2′); 70.32 (d, $J_{C,P}$ = 6, $CH(CH_3)_2$); 107.11 (CH-7); 112.29 (C-9a); 119.64 (CH-6); 142.98 (C-9); 143.80 (CH-2); 143.91 (C-4a); 151.21 (C-3a).

5.11. Diisopropyl (2-(9-(dimethylamino)-3*H*-imidazo[1,2-*a*]purin-3-yl)ethoxy)methylphosphonate (9c) and diisopropyl (2-(4-(dimethylamino)-1*H*-imidazo[2,1-*b*]purin-1-yl)ethoxy)methylphosphonate (10c)

Prepared from **5c** (1.2 g, 3 mmol) and chloroacetaldehyde (20 ml, 1 M aq solution). Chromatography over silica gel in MeOH/$CHCl_3$ 4:96 yielded 0.36 g (28%) of **9c** as yellow syrupy product followed by 0.62 g (49%) of **10c** as light brown syrupy product. **9c**: MS (FAB) *m*/*z* (rel intensity): 425 (100, M+1). HR MS (FAB): for $C_{18}H_{29}N_6O_4P$ calcd 425.2066, found 425.2060. UV (MeOH): 343 (3300), 284 (5980), 242 (16270). FT IR (KBr): 3432, 2956, 2922, 2853, 1685, 1654, 1631, 1584, 1541, 1492, 1466, 1400, 1388, 1375, 1250, 1180, 1106,

1081, 984, 889. ^{1}H NMR (500 MHz, DMSO-d_6): 1.11 and 1.15 (2× d, 2× 6H, J_{vic} = 6.2, (CH_3)$_2$CH); 3.36 (s, 6H, (CH$_3$)$_2$N); 3.79 (d, 2H, $J_{H,P}$ = 8.3, H-4′); 3.92 (t, 2H, $J_{2',1'}$ = 5.2, H-2′); 4.32 (t, 2H, $J_{1',2'}$ = 5.2, H-1′); 4.49 (dh, 2H, $J_{H,P}$ = 7.7, J_{vic} = 6.2, CH(CH$_3$)$_2$); 7.51 (d, 1H, $J_{6,7}$ = 1.8, H-6); 7.73 (d, 1H, $J_{7,6}$ = 1.8, H-7); 8.17 (s, 1H, H-2). ^{13}C NMR (125.8 MHz, DMSO-d_6): 23.70 and 23.85 (d, $J_{C,P}$ = 4, (CH$_3$)$_2$CH); 41.78 ((CH$_3$)$_2$N); 42.31 (CH$_2$-1′); 64.70 (d, $J_{C,P}$ = 164, CH$_2$-4′); 70.01 (d, $J_{C,P}$ = 12, CH$_2$-2′); 70.29 (d, $J_{C,P}$ = 6, CH(CH$_3$)$_2$); 107.83 (CH-7); 115.58 (C-9a); 132.38 (CH-6); 143.69 (C-9); 144.73 (CH-2); 149.42 (C-4a); 150.96 (C-3a). **10c**: MS (FAB) *m*/*z* (rel intensity): 425 (100, M+1). HR MS (FAB): for $C_{18}H_{29}N_6O_4P$ calcd 425.2066, found 425.2062. UV (MeOH): 287 (15200), 238 (31620). FT IR (KBr): 3423, 3086, 2979, 2932, 2878, 1649, 1634, 1584, 1467, 1421, 1407, 1387, 1237, 1178, 1165, 1142, 1107, 987, 890. ^{1}H NMR (500 MHz, DMSO-d_6): 1.06 and 1.10 (2× d, 2× 6H, J_{vic} = 6.2, (CH_3)$_2$CH); 3.46 (br s, 6H, (CH$_3$)$_2$N); 3.75 (d, 2H, $J_{H,P}$ = 7.9, H-4′); 3.92 (t, 2H, $J_{2',1'}$ = 5.1, H-2′); 4.39 (dh, 2H, $J_{H,P}$ = 7.7, J_{vic} = 6.2, CH(CH$_3$)$_2$); 4.71 (t, 2H, $J_{1',2'}$ = 5.2, H-1′); 7.35 (d, 1H, $J_{7,8}$ = 1.9, H-7); 7.92 (d, 1H, $J_{8,7}$ = 1.9, H-8); 8.01 (s, 1H, H-2). ^{13}C NMR (125.8 MHz, DMSO-d_6): 23.66 and 23.78 (d, $J_{C,P}$ = 4, (CH$_3$)$_2$CH); 38.71 ((CH$_3$)$_2$N); 45.39 (CH$_2$-1′); 64.96 (d, $J_{C,P}$ = 163, CH$_2$-4′); 70.23 (d, $J_{C,P}$ = 6, CH(CH$_3$)$_2$); 71.12 (d, $J_{C,P}$ = 10, CH$_2$-2′); 106.89 (CH-8); 116.44 (C-3a); 128.33 (CH-7); 134.25 (C-9a); 137.65 (CH-2); 147.47 (C-5a); 150.93 (C-4).

5.12. Diisopropyl (2-(9-(pyrrolidin-1-yl)-3*H*-imidazo[1,2-*a*]purin-3-yl)ethoxy)methylphosphonate (9d) and Diisopropyl (2-(4-(pyrrolidin-1-yl)-1*H*-imidazo[2,1-*b*]purin-1-yl)ethoxy)methylphosphonate (10d)

Prepared from **5d** (1.1 g, 2.6 mmol) and chloroacetaldehyde (20 ml, 1 M aq solution). Chromatography over silica gel in MeOH/CHCl$_3$ 5:95 afforded 0.25 g (22%) of **9d** followed by 0.42 g (36%) of **10d** as light brown syrupy products. **9d**: MS (FAB) *m*/*z* (rel intensity): 451 (100, M+1). HR MS (FAB): for $C_{20}H_{31}N_6O_4P$ calcd 451.2222, found 451.2202. UV (MeOH): 332 (3700), 281 (7660), 238 (16820). FT IR (KBr): 3422, 2978, 2933, 2876, 1648, 1596, 1554, 1540, 1458, 1686, 1375, 1353, 1243, 1178, 1106, 990, 890. ^{1}H NMR (400 MHz, DMSO-d_6): 1.12 and 1.16 (2× d, 2× 6H, J_{vic} = 6.2, (C**H**$_3$)$_2$CH); 1.98 (m, 4H, CH$_2$-pyrrolidine); 3.79 (d, 2H, $J_{H,P}$ = 8.4, H-4′); 3.89 (t, 2H, $J_{2',1'}$ = 5.2, H-2′); 4.20 (m, 4H, CH$_2$N-pyrrolidine); 4.27 (t, 2H, $J_{1',2'}$ = 5.2, H-1′); 4.50 (dh, 2H, $J_{H,P}$ = 7.7, J_{vic} = 6.2, CH(CH$_3$)$_2$); 7.39 (d, 1H, $J_{6,7}$ = 1.9, H-6); 7.96 (d, 1H, $J_{7,6}$ = 1.9, H-7); 8.02 (s, 1H, H-2). ^{13}C NMR (100.6 MHz, DMSO-d_6): 23.77 and 23.91 (d, $J_{C,P}$ = 4, (CH$_3$)$_2$CH); 25.60 (CH$_2$-pyrrolidine); 42.20 (CH$_2$-1′); 52.34 (CH$_2$N-pyrrolidine); 64.71 (d, $J_{C,P}$ = 164, CH$_2$-4′); 70.10 (d, $J_{C,P}$ = 12, CH$_2$-2′); 70.34 (d, $J_{C,P}$ = 6, CH(CH$_3$)$_2$); 108.56 (CH-7); 112.98 (C-9a); 130.81 (CH-6); 142.32 (C-9); 142.44 (CH-2); 150.11 (C-4a); 150.59 (C-3a). **10d**: MS (FAB) *m*/*z* (rel intensity): 451 (100, M+1). HR MS (FAB): for $C_{20}H_{31}N_6O_4P$ calcd 451.2222, found 451.2224. UV (MeOH): 288 (7360), 238 (13600). FT IR (KBr): 3430, 2987, 2933, 2877, 1647, 1583, 1454, 1386, 1375, 1349, 1233, 1178, 1142, 1106, 990, 891. ^{1}H NMR (400 MHz, DMSO-d_6): 1.06 and 1.10 (2× d, 2× 6H, J_{vic} = 6.2, (CH_3)$_2$CH); 1.98 (br m, 4H, CH$_2$-pyrrolidine); 3.73 (d, 2H, $J_{H,P}$ = 8.0, H-4′); 3.91 (t, 2H, $J_{2',1'}$ = 5.2, H-2′); 3.9-4.3 (br m, 4H, CH$_2$N-pyrrolidine); 4.38 (dh, 2H, $J_{H,P}$ = 7.7, J_{vic} = 6.2, CH(CH$_3$)$_2$); 4.76 (t, 2H, $J_{1',2'}$ = 5.2, H-1′); 7.56 (d, 1H, $J_{7,8}$ = 1.9, H-7); 8.06 (d, 1H, $J_{8,7}$ = 1.9, H-8); 8.13 (s, 1H, H-2). ^{13}C NMR (100.6 MHz, DMSO-d_6): 23.70 and 23.81 (d, $J_{C,P}$ = 4, (CH$_3$)$_2$CH); 25.58 (CH$_2$-pyrrolidine); 45.57 (CH$_2$-1′); 52.88 (CH$_2$N-pyrrolidine); 64.97 (d, $J_{C,P}$ = 164, CH$_2$-4′); 70.21 (d, $J_{C,P}$ = 6, CH(CH$_3$)$_2$); 71.17 (d, $J_{C,P}$ = 11, CH$_2$-2′); 108.32 (CH-8); 117.12 (C-3a); 122.94 (CH-7); 134.00 (C-9a); 139.11 (CH-2); 145.61 (C-5a); 149.95 (C-4).

5.13. Diisopropyl (2-(9-amino-3*H*-imidazo[1,2-*a*]purin-3-yl)ethoxy)methoxyphosphonate (9e) and Diisopropyl (2-(4-amino-1*H*-imidazo[2,1-*b*]purin-1-yl)ethoxy)methylphosphonate (10e)

Prepared from **5e** (370 mg, 1 mmol) and chloroacetaldehyde (15 ml, 1 M aq solution). Chromatography over silica gel in MeOH/CHCl$_3$ 5:95 afforded 60 mg (15%) of **9e** as yellow syrupy product followed by 170 mg (43%) of **10e** as light brown syrupy product. **9e**: MS (FAB) *m*/*z* (rel intensity): 397 (100, M+1). HR MS (FAB): for $C_{16}H_{25}N_6O_4P$ calcd 397.1753, found 397.1757. UV (MeOH): 278 (7800), 218 (18160). FT IR (KBr): 3429, 2960, 2920, 2852, 1654, 1637, 1559, 1491, 1468, 1399, 1388, 1362, 1213, 1196, 1082, 991, 854. ^{1}H NMR (400 MHz, DMSO-d_6): 1.11 and 1.16 (2× d, 2× 6H, J_{vic} = 6.2, (CH_3)$_2$CH); 3.78 (d, 2H, $J_{H,P}$ = 8.4, H-4′); 3.90 (t, 2H, $J_{2',1'}$ = 5.2, H-2′); 4.30 (t, 2H, $J_{1',2'}$ = 5.2, H-1′); 4.49 (dh, 2H, $J_{H,P}$ = 7.7, J_{vic} = 6.2, CH(CH$_3$)$_2$); 7.41 (d, 1H, $J_{6,7}$ = 1.6, H-6); 7.55 (br s, 2H, NH$_2$); 7.85 (s, 1H, H-2); 7.94 (d, 1H, $J_{7,6}$ = 1.6, H-7). ^{13}C NMR (100.6 MHz, DMSO-d_6): 23.76 and 23.90 (d, $J_{C,P}$ = 4, (CH$_3$)$_2$CH); 42.77 (CH$_2$-1′); 64.79 (d, $J_{C,P}$ = 164, CH$_2$-4′); 70.35 (d, $J_{C,P}$ = 6, CH(CH$_3$)$_2$); 70.68 (d, $J_{C,P}$ = 12, CH$_2$-2′); 109.65 (CH-7); 115.58 (C-9a); 131.26 (CH-6); 137.75 (CH-2); 141.26 (C-3a); 142.25 (C-4a); 145.06 (C-9). **10e**: MS (FAB) *m*/*z* (rel intensity): 397 (100, M+1). HR MS (FAB): for $C_{16}H_{25}N_6O_4P$ calcd 397.1753, found 397.1743. UV (MeOH): 324 (4470), 274 (3700), 237 (25900). FT IR (KBr): 3429, 3104, 2980, 2925, 2854, 1665, 1633, 1584, 1546, 1524, 1451, 1409, 1387, 1375, 1227, 1179, 1143, 1106, 999. ^{1}H NMR (500 MHz, DMSO-d_6): 1.10 and 1.15 (2× d, 2× 6H, J_{vic} = 6.2, (CH_3)$_2$CH); 3.79 (d, 2H, $J_{H,P}$ = 8.0, H-4′); 3.91 (t, 2H, $J_{2',1'}$ = 5.1, H-2′); 4.30 (t, 2H, $J_{1',2'}$ = 5.1, H-1′); 4.49 (dh, 2H, $J_{H,P}$ = 7.6, J_{vic} = 6.2, CH(CH$_3$)$_2$); 7.51 (d, 1H, $J_{7,8}$ = 1.9, H-7); 8.00 (d, 1H, $J_{8,7}$ = 1.9, H-8); 8.09 (s, 1H, H-2); 8.63 (br s, 2H, NH$_2$). ^{13}C NMR (125.8 MHz, DMSO-d_6): 23.77 and 23.92 (d, $J_{C,P}$ = 4, (CH$_3$)$_2$CH); 42.40 (CH$_2$-1′); 64.69 (d, $J_{C,P}$ = 164, CH$_2$-4′); 71.23 (d, $J_{C,P}$ = 12, CH$_2$-2′); 70.35 (d, $J_{C,P}$ = 6, CH(CH$_3$)$_2$); 105.74 (CH-8); 109.87 (C-3a); 129.54 (CH-7); 141.92 (C-9a); 142.96 (CH-2); 148.11 (C-5a); 149.86 (C-4).

5.14. Diisopropyl (2-(4-(diethylamino)-1*H*-imidazo[2,1-*b*]purin-1-yl)ethoxy)methylphosphonate (10b)

Prepared from **5b** (2 g, 4.7 mmol) and chloroacetaldehyde (30 ml, 1 M aq solution). Chromatography over silica gel in MeOH/$CHCl_3$ 5:95 afforded 1 g (48%) of **10b** as yellow syrupy product. MS (FAB) *m/z* (rel intensity): 453 (100, M+1). HR MS (FAB): for $C_{20}H_{33}N_6O_4P$ calcd 453.2379, found 453.2396. UV (MeOH): 283 (12600), 236 (18600). FT IR (KBr): 3428, 2979, 2932, 2875, 1647, 1594, 1579, 1507, 1465, 1438, 1386, 1377, 1329, 1254, 1233, 1178, 1105, 994, 891. ^{1}H NMR (400 MHz, DMSO-d_6): 1.06 and 1.08 (2× d, 2× 6H, J_{vic} = 6.2, $(CH_3)_2CH$); 1.23 (t, 6H, J_{vic} = 6.9, CH_3CH_2); 3.74 (d, 2H, $J_{H,P}$ = 8.0, H-4′); 3.91 (t, 2H, $J_{2',1'}$ = 5.3, H-2′); 3.99 (br m, 4H, CH_2CH_3); 4.36 (dh, 2H, $J_{H,P}$ = 7.7, J_{vic} = 6.2, $CH(CH_3)_2$); 4.73 (t, 2H, $J_{1',2'}$ = 5.3, H-1′); 7.48 (d, 1H, $J_{7,8}$ = 2.1, H-7); 7.99 (d, 1H, $J_{8,7}$ = 2.1, H-8); 8.09 (s, 1H, H-2). ^{13}C NMR (100.6 MHz, DMSO-d_6): 13.63 (CH_3CH_2); 23.67 and 23.79 (d, $J_{C,P}$ = 4, $(CH_3)_2CH$); 43.21 (CH_2CH_3); 45.53 (CH_2-1′); 64.94 (d, $J_{C,P}$ = 163, CH_2-4′); 70.20 (d, $J_{C,P}$ = 6, $CH(CH_3)_2$); 71.12 (d, $J_{C,P}$ = 11, CH_2-2′); 107.80 (CH-8); 116.33 (C-3a); 124.94 (CH-7); 134.38 (C-9a); 138.58 (CH-2); 146.27 (C-5a); 150.15 (C-4).

5.15. Cleavage of protecting isopropyl esters. General procedure

Compounds **7**, **9a**–**e**, and **10a**–**e** were dissolved in dry acetonitrile (70 ml). Bromotrimethylsilane (2 ml) was added and the reaction mixture was stirred at ambient temperature for 24 h. The solvent was removed by evaporation in vacuo, the residue was treated with water, and neutralized with ammonia solution. The mixture was evaporated in vacuo, the residue dissolved in minimum amount of water, and applied on a Dowex 50 × 8 column in H^+ cycle. The column was washed with water followed by a solution of ammonia/water 1:10 and the fractions with UV absorption were collected, evaporated in vacuo, the residue was dissolved in minimum amount of water and alkalized with ammonia. Then it was applied on a Dowex 1 × 2 column in AcO^- cycle and washed with water followed by linear gradient of acetic acid solution (0.01–0.5 M). Fractions containing product were collected, the solvent was removed in vacuo, and the residue was codistilled with methanol. Then it was dissolved in minimum amount of methanol and the product crystallized upon addition of excessive amount of diethyl ether. The product was filtered off and dried. This procedure was used for the following compounds.

5.16. Sodium (2-(9-oxo-5,9-dihydro-3*H*-imidazo[1,2-*a*]purin-3-yl)ethoxy)methylphosphonate (8)

Prepared from **7** (120 mg, 0.3 mmol), afforded 40 mg (42%) of **8** as slightly colored powder, mp >300 °C. Instead of Dowex 1 column, DEAE Sephadex column was used and the product was eluted with gradient of tetrabutylammoniumbicarbonate (0.0–0.6 M). Then it was treated with Dowex 50 in Na^+ cycle, filtered off, and evaporated in vacuo to give product in the form of the corresponding sodium salt. MS (FAB) *m/z* (rel intensity): 314 (72, M+1). UV (H_2O): 282 (4320), 224 (12350). FT IR (KBr): 3425, 3103, 2924, 2854, 2759, 1685, 1600, 1572, 1549, 1533, 1454, 1391, 1293, 1218, 1107, 1067, 912. ^{1}H NMR (400 MHz, D_2O + NaOD, ref(dioxane) = 3.75 ppm): 3.51 (d, 2H, $J_{H,P}$ = 8.5, H-4′); 3.96 (t, 2H, $J_{2',1'}$ = 5.0, H-2′); 4.32 (t, 2H, $J_{1',2'}$ = 5.0, H-1′); 7.26 (d, 1H, $J_{6,7}$ = 1.7, H-6); 7.59 (d, 1H, $J_{7,6}$ = 1.7, H-7); 7.98 (s, 1H, H-2). ^{13}C NMR (100.6 MHz, D_2O + NaOD, ref(dioxane) = 69.3 ppm): 45.60 (CH_2-1′); 71.70 (d, $J_{C,P}$ = 150, CH_2-4′); 72.88 (d, $J_{C,P}$ = 10, CH_2-2′); 108.72 (CH-7); 115.31 (C-9a); 131.29 (CH-6); 144.09 (CH-2); 153.72 (C-3a); 154.16 (C-4a); 157.11 (C-9). For $C_{10}H_{10}N_5Na_2O_5P$ calcd 33.63% C, 2.82% H, 19.61% N, 12.87% Na, 8.67% P, found 33.31% C, 2.88% H, 19.43% N, 8.51% P.

5.17. (2-(9-(Cyclopropylimino)-5,9-dihydro-3*H*-imidazo[1,2-*a*]purin-3-yl)ethoxy)methylphosphonic acid (11a)

Prepared from **9a** (340 mg, 0.8 mmol), afforded 60 mg (22%) of **11a** as light brown powder, mp >300 °C. MS (FAB) *m/z* (rel intensity): 353 (90, M+1). UV (H_2O): 325 (4830), 285 sh (2700), 277 (2730 sh), 239 (17300). FT IR (KBr): 3412, 3146, 3050, 2836, 1663, 1589, 1535, 1514, 1407, 1286, 1172, 1121, 1053, 971. ^{1}H NMR (500 MHz, D_2O, ref(dioxane) = 3.75 ppm): 0.83 and 1.03 (2× m, 2× 2H, CH_2-cycloprop); 3.32 (tt, 1H, J = 7.1, 3.7, CH-N); 3.63 (d, 2H, $J_{H,P}$ = 8.8, H-4′); 3.98 (t, 2H, $J_{2',1'}$ = 4.9, H-2′); 4.35 (t, 2H, $J_{1',2'}$ = 4.9, H-1′); 7.50 (d, 1H, $J_{6,7}$ = 2.5, H-6); 7.64 (d, 1H, $J_{7,6}$ = 2.5, H-7); 8.17 (s, 1H, H-2). ^{13}C NMR (125.8 MHz, D_2O, ref(dioxane) = 69.3 ppm): 10.58 (CH_2-cycloprop); 29.55 (CH–N); 46.00 (CH_2-1′); 70.71 (d, $J_{C,P}$ = 153, CH_2-4′); 72.69 (d, $J_{C,P}$ = 12, CH_2-2′); 109.48 (CH-7); 113.39 (C-9a); 125.59 (CH-6); 145.26 (C-9); 147.24 (CH-2); 147.36 (C-4a); 153.36 (C-3a). For $C_{13}H_{17}N_6O_4P$ calcd 44.32% C, 4.86% H, 23.86% N, 8.79% P, found 44.05% C, 4.54%H, 23.48% N, 8.60% P.

5.18. (2-(9-(Dimethylamino)-3*H*-imidazo[1,2-*a*]purin-3-yl)ethoxy)methylphosphonic acid (11c)

Prepared from **9c** (360 mg, 0.8 mmol), afforded 120 mg (41%) of **11c** as white powder, mp 250–254 °C. MS (FAB) *m/z* (rel intensity): 341 (40, M+1), 316 (20), 288 (53). UV (H_2O): 345 (6600), 294 (5720), 289 (5700), 244 (24350). FT IR (KBr): 3141, 3046, 2806, 1656, 1578, 1442, 1404, 0089, 1128, 997. ^{1}H NMR (500 MHz, D_2O + NaOD, ref(dioxane) = 3.75 ppm): 3.35 (s, 6H, $(CH_3)_2N$); 3.48 (d, 2H, $J_{H,P}$ = 8.5, H-4′); 3.92 (t, 2H, $J_{2',1'}$ = 5.1, H-2′); 4.29 (t, 2H, $J_{1',2'}$ = 5.1, H-1′); 7.39 (d, 1H, $J_{6,7}$ = 2.0, H-6); 7.58 (d, 1H, $J_{7,6}$ = 2.0, H-7); 8.12 (s, 1H, H-2). ^{13}C NMR (125.8 MHz, D_2O + NaOD, ref(dioxane) = 69.3 ppm): 44.29 ($(CH_3)_2N$); 45.71 (CH_2-1′); 71.76 (d, $J_{C,P}$ = 150, CH_2-4′); 72.52 (d, $J_{C,P}$ = 10, CH_2-2′); 111.93 (CH-7); 116.88 (C-9a); 133.46 (CH-6); 147.34 (C-9); 147.38 (CH-2); 152.22 (C-4a); 153.11 (C-3a). For $C_{12}H_{17}N_6O_4P$ calcd 42.36% C, 5.04% H, 24.70% N, 9.10% P, found 42.02% C, 5.36% H, 24.32% N, 8.84% P.

5.19. (2-(9-(Pyrrolidin-1-yl)-3*H*-imidazo[1,2-*a*]purin-3-yl)ethoxy)methylphosphonic acid (11d)

Prepared from **9d** (250 mg, 0.5 mmol), afforded 65 mg (32%) of **11d** as white amorphous powder. MS (FAB) *m/z* (rel intensity): 367 (100, M+1), 343 (37). UV (H_2O): 342 (5850), 283 (5550), 245 (21600). FT IR (KBr): 3113, 2924, 2883, 2747, 1654, 1579, 1556, 1453, 1407, 1347, 1219, 1119, 990, 927. ^{1}H NMR (500 MHz, D_2O, ref(dioxane) = 3.75 ppm): 2.10 (m, 4H, CH_2-pyrrolidine); 3.56 (d, 2H, $J_{H,P}$ = 8.7, H-4′); 3.95 (t, 2H, $J_{2',1'} = 4.9$, H-2′); 4.22 (m, 4H, CH_2N-pyrrolidine); 4.32 (t, 2H, $J_{1',2'} = 4.9$, H-1′); 7.45 (d, 1H, $J_{6,7}$ = 2.7, H-6); 8.03 (d, 1H, $J_{7,6}$ = 2.7, H-7); 8.11 (s, 1H, H-2). ^{13}C NMR (125.8 MHz, D_2O, ref(dioxane) = 69.3 ppm): 28.10 (CH_2-pyrrolidine); 45.89 (CH_2-1′); 56.62 (CH_2N-pyrrolidine); 70.87 (d, $J_{C,P}$ = 153, CH_2-4′); 72.72 (d, $J_{C,P}$ = 11, CH_2-2′); 113.45 (CH-7); 115.98 (C-9a); 123.57 (CH-6); 145.74 (CH-2); 145.93 (C-9); 149.37 (C-4a); 152.63 (C-3a). For $C_{14}H_{19}N_6O_4P$ calcd 45.90% C, 5.23% H, 22.94% N, 8.46% P, found 45.55% C, 5.48% H, 22.61% N, 8.10% P.

5.20. (2-(9-Amino-3*H*-imidazo[1,2-*a*]purin-3-yl)ethoxy)methylphosphonic acid (11e)

Prepared from **9e** (120 mg, 0.3 mmol), afforded 35 mg (37%) of **11e** as slightly colored powder, mp 185–190 °C. MS (FAB) *m/z* (rel intensity): 313 (67, M+1), 289 (100). UV (H_2O): 308 (2400 sh), 280 (7700), 254 (5300), 215 (17630). FT IR (KBr): 3407, 3142, 3049, 2925, 1692, 1650, 1581, 1531, 1407, 1192, 1110, 990, 932. ^{1}H NMR (400 MHz, D_2O + NaOD, ref(dioxane) = 3.75 ppm): 3.50 (d, 2H, $J_{H,P}$ = 8.5, H-4′); 3.29 (t, 2H, $J_{2',1'} = 5.3$, H-2′); 4.23 (t, 2H, $J_{1',2'} = 5.3$, H-1′); 7.30 (d, 1H, $J_{6,7}$ = 1.7, H-6); 7.46 (d, 1H, $J_{7,6}$ = 1.7, H-7); 7.88 (s, 1H, H-2). ^{13}C NMR (100.6 MHz, D_2O + NaOD, ref(dioxane) = 69.3 ppm): 46.01 (CH_2-1′); 71.66 (d, $J_{C,P}$ = 150, CH_2-4′); 72.97 (d, $J_{C,P}$ = 10, CH_2-2′); 112.39 (CH-7); 116.99 (C-9a); 133.41 (CH-6); 141.63 (CH-2); 142.92 (C-3a); 144.14 (C-4a); 147.70 (C-9). For $C_{10}H_{13}N_6O_4P$ calcd 38.47% C, 4.20% H, 26.92% N, 9.92% P, found 38.20% C, 4.52% H, 26.58% N, 9.68% P.

5.21. (2-(4-(Diethylamino)-1-*H*-imidazo[2,1-*b*]purin-1-yl)ethoxy)methylphosphonic acid (12b)

Prepared from **10b** (1 g, 2 mmol), afforded 240 mg (28%) of **12b** as white amorphous powder. MS (FAB) *m/z* (rel intensity): 369 (100, M+1). UV (H_2O): 287 (16300), 251 (16700, sh), 237 (29100). FT IR (KBr): 3420, 2975, 2935, 2894, 2361, 1653, 1577, 1507, 1464, 1438, 1381, 1363, 1329, 1254, 1235, 1176, 1114, 1079, 989, 932. ^{1}H NMR (400 MHz, D_2O, ref(dioxane) = 3.75 ppm): 1.23 (br t, 6H, J_{vic} = 6.7, CH_3CH_2); 3.56–3.79 and 3.91–4.16 (2× vb, 2× 2H, CH_2CH_3); 3.68 (d, 2H, $J_{H,P}$ = 8.8, H-4′); 4.04 (t, 2H, $J_{2',1'} = 5.0$, H-2′); 4.73 (t, 2H, $J_{1',2'} = 5.0$, H-1′); 7.43 (d, 1H, $J_{7,8}$ = 2.7, H-7); 7.91 (d, 1H, $J_{8,7}$ = 2.7, H-8); 8.09 (s, 1H, H-2). ^{13}C NMR (100.6 MHz, D_2O, ref(dioxane) = 69.3 ppm): 16.12 (CH_3CH_2); 47.20 (CH_2CH_3); 49.11 (CH_2-1′); 69.02 (d, $J_{C,P}$ = 159, CH_2-4′); 73.79 (d, $J_{C,P}$ = 12, CH_2-2′); 111.56 (CH-8); 119.43 (C-3a); 120.16 (CH-7); 136.74 (C-9a); 142.23 (CH-2); 145.96 (C-5a); 153.99 (C-4). For $C_{14}H_{21}N_6O_4P$ calcd 45.65% C, 5.75% H, 22.82% N, 8.41% P, found 45.28% C, 6.09% H, 22.65% N, 8.13% P.

5.22. (2-(4-(Dimethylamino)-1-*H*-imidazo[2,1-*b*]purin-1-yl)ethoxy)methylphosphonic acid (12c)

Prepared from **10c** (500 mg, 1.2 mmol), afforded 130 mg (32%) of **12c** as white powder, mp 180–185 °C. MS (FAB) *m/z* (rel intensity): 341 (60, M+1). UV (H_2O): 284 (12500), 251 (12700 sh), 236 (23100). FT IR (KBr): 3145, 3050, 2815, 1654, 1588, 1549, 1509, 1407, 1232, 1113, 977. ^{1}H NMR (400 MHz, D_2O, ref(dioxane) = 3.75 ppm): 3.32 (br s, 6H, $(CH_3)_2N$); 3.44 (d, 2H, $J_{H,P}$ = 8.4, H-4′); 3.96 (t, 2H, $J_{2',1'} = 5.2$, H-2′); 4.55 (t, 2H, $J_{1',2'} = 5.2$, H-1′); 7.25 (d, 1H, $J_{7,8}$ = 1.9, H-7); 7.63 (d, 1H, $J_{8,7}$ = 1.9, H-8); 7.91 (s, 1H, H-2). ^{13}C NMR (125.8 MHz, D_2O+NaOD, ref(dioxane) = 69.3 ppm): 41.72 ($(CH_3)_2N$); 48.43 (CH_2-1′); 71.87 (d, $J_{C,P}$ = 150, CH_2-4′); 72.78 (d, $J_{C,P}$ = 10, CH_2-2′); 109.24 (CH-8); 118.13 (C-3a); 132.18 (CH-7); 136.48 (C-9a); 140.36 (CH-2); 151.08 (C-5a); 153.55 (C-4). For $C_{12}H_{17}N_6O_4P$ calcd 42.36% C, 5.04% H, 24.70% N, 9.10% P, found 42.20% C, 5.28% H, 24.52% N, 8.90% P.

5.23. (2-(4-(Pyrrolidin-1-yl)-1-*H*-imidazo[2,1-*b*]purin-1-yl)ethoxy)methylphosphonic acid (12d)

Prepared from **10d** (420 mg, 0.9 mmol), afforded 105 mg (31%) of **12d** as slightly colored powder, mp 175–180 °C. MS (FAB) *m/z* (rel intensity): 367 (100, M+1). UV (H_2O): 283 (10160), 251 (12700 sh), 16350. FT IR (KBr): 3417, 3143, 3051, 2880, 2742, 1651, 1584, 1506, 1454, 1409, 1349, 1227, 1212, 1114, 994, 933. ^{1}H NMR (400 MHz, DMSO-d_6): 1.95 and 2.09 (2× p, 2× 2H, J_{vic} = 7.0, CH_2-pyrrolidine); 3.45 (t, 2H, J_{vic} = 7.0, CH_2N-pyrrolidine); 3.62 (d, 2H, $J_{H,P}$ = 8.7, H-4′); 3.97 (t, 2H, J_{vic} = 7.0, CH_2N-pyrrolidine); 4.04 (t, 2H, $J_{2',1'} = 5.0$, H-2′); 4.70 (t, 2H, $J_{1',2'} = 5.0$, H-1′); 7.40 (d, 1H, $J_{7,8}$ = 2.7, H-7); 7.92 (d, 1H, $J_{8,7}$ = 2.7, H-8); 8.12 (s, 1H, H-2). ^{13}C NMR (100.6 MHz, DMSO-d_6): 26.12 and 28.52 (CH_2-pyrrolidine); 49.22 (CH_2-1′); 51.51 and 52.39 (CH_2N-pyrrolidine); 69.99 (d, $J_{C,P}$ = 157, CH_2-4′); 73.53 (d, $J_{C,P}$ = 11, CH_2-2′); 111.57 (CH-8); 119.71 (C-3a); 120.11 (CH-7); 136.27 (C-9a); 142.57 (CH-2); 145.96 (C-5a); 152.91 (C-4). For $C_{14}H_{19}N_6O_4P$ calcd 45.90% C, 5.23% H, 22.94% N, 8.46% P, found 45.60% C, 5.56% H, 22.72% N, 8.25% P.

5.24. (2-(4-Amino-1*H*-imidazo[2,1-*b*]purin-1-yl)ethoxy)methylphosphonic acid (12e)

Prepared from **10e** (170 mg, 0.4 mmol), afforded 35 mg (27%) of **12e** as slightly colored powder, mp 270–280 °C. MS (FAB) *m/z* (rel intensity): 313 (52, M+1). UV (H_2O): 324 (3500), 308 (3250), 274 (3030), 237 (19500). FT IR (KBr): 3397, 3132, 3050, 1688, 1583, 1560, 1526, 1446, 1408, 1324, 1227, 1145, 1117, 1054, 916. ^{1}H NMR (400 MHz, D_2O + NaOD, ref(dioxane) = 3.75 ppm): 3.50 (d, 2H, $J_{H,P}$ = 8.5, H-4′); 3.93 (t, 2H, $J_{2',1'} = 5.3$, H-2′); 4.26 (t, 2H, $J_{1',2'} = 5.3$, H-1′); 7.25 (d, 1H, $J_{7,8}$ = 1.9, H-7); 7.50 (d, 1H, $J_{8,7}$ = 1.9,

H-8); 7.90 (s, 1H, H-2). ^{13}C NMR (100.6 MHz, D_2O+NaOD, ref(dioxane) = 69.3 ppm): 45.55 (CH_2-1′); 71.71 (d, $J_{C,P} = 150$, CH_2-4′); 72.87 (d, $J_{C,P} = 10$, CH_2-2′); 108.85 (CH-8); 113.15 (C-3a); 131.62 (CH-7); 143.09 (CH-2); 150.11 (C-9a); 152.14 (C-4); 153.35 (C-5a). For $C_{10}H_{13}N_6O_4P$ calcd 38.47% C, 4.20% H, 26.92% N, 9.92% P, found 38.05% C, 4.61% H, 26.64% N, 9.75% P.

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