Synthesis and Cytostatic Activity of Substituted 6-Phenylpurine Bases and Nucleosides: Application of the Suzuki−Miyaura Cross-Coupling Reactions of 6-Chloropurine Derivatives with Phenylboronic Acids

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Received September 24, 1999

The Suzuki−Miyaura reaction of protected 6-chloropurine and 2-amino-6-chloropurine bases and nucleosides with substituted phenylboronic acids led to the corresponding protected 6-(substituted phenyl)purine derivatives **6**−**9**. Their deprotection yielded a series of substituted 6-phenylpurine bases and nucleosides **10**−**13**. Significant cytostatic activity (IC_{50} 0.25−20 μmol/L) in CCRF-CEM, HeLa, and L1210 cell lines was found for several 6-(4-X-substituted phenyl)-purine ribonucleosides **12** (X = H, F, Cl, and OR), while the 6-phenylpurine and 2-amino-6-phenylpurine bases **10** and **11**, as well as 2-amino-6-phenylpurine ribosides **13,** were entirely inactive against these cell lines.

Introduction

Purine bases and their nucleosides constitute an important group of antineoplastic and antileukemic agents (for reviews, see ref 1). Thus, olomoucin and its congeners exhibit their cytostatic activity by virtue of inhibiting the cyclin-dependent protein kinases.[2] In the search for novel antimetabolites, purine bases as well as the carbohydrate residue in the nucleoside molecules were modified by structural alterations. An exhaustive review covers numerous cytostatic aza and deaza analogues of purine nucleosides.[3−5] The most attractive compound of this group is 3-deazaguanine (NSC 261726)[6] and its ribonucleoside,[7] but an important antineoplastic activity was also encountered among 7-deazapurine nucleoside antibiotics (tubercidin, toyocamycin, sangivamycin) and their sugar-modified analogues.[3,8]

Replacement of the oxygen atom at position 6 in guanine and hypoxanthine by sulfur gave clinically widely used anticancer drugs thioguanine (NSC752)[9] and 6-mercaptopurine (NSC755).[10] Structurally related nucleoside sulfinosine [2-amino-9-(β-D-ribofuranosyl)-purine-6-sulfinamide] and its congeners[11] also exhibit unique antitumor properties.

Substitution at position 2 of the adenine ring by a halogen atom gives rise to biologically active nucleosides: while 2-chloroadenosine (an adenosine receptor agonist) causes solely a selective depletion of natural killer cells via signal transduction,[12] substantial therapeutic success was achieved with 2-chloro-2′-deoxyadenosine[13] (NSC 105014-F, cladribine) which is clinically used in the treatment of hairy cell leukemia, human primary lymphoma, chronic lymphocytic leukemia, and myeloid leukemia.[14] A closely related drug in clinical use for treatment of lymphoproliferative diseases is 9-(β-D-arabinofuranosyl)-2-fluoroadenine (fludarabine) and its water-soluble 5′-phosphate (NSC 312887, fludarabine phosphate).[15] Also the influence of introduction of alkyl substituents at position 2 of the purine bases has been examined, but no cytostatic activity was noted among these compounds; only the antihypertensive activity of 2-octynyladenosine was reported.[16]

Most of the above substitutions maintain the amino or oxo (thioxo) group at position 6 of the purine base which is essential for the formation of hydrogen bonds important for the interactions with polymerases and other key enzymes of nucleic acid metabolism. The cytostatic activity was also observed in 2-amino-6-methoxypurine arabinoside, which was investigated as a possible tool for combating T-cell malignancies,[17] and the 6-alkylmercaptopurine derivatives exhibiting certain cytostatic activity;[18] nonetheless, these compounds might be eventually converted to guanine nucleosides by the action of adenosine aminohydrolase.

Such a catabolic reaction is indeed excluded in 6-alkylpurine derivatives. The parent compound of this group, 6-methylpurine, is known for its cytotoxicity; its liberation from the 2′-deoxyribonucleoside by purine nucleoside phosphorylases is used for detection of mycoplasma in cell cultures.[19] It is highly potent and toxic to nonproliferating and proliferating tumor cells. Recently, the use of cytotoxic bases liberated by purine nucleoside phosphorylases such as 6-methylpurine was proposed as a novel principle in the gene therapy of cancer.[20]

Peculiarly little attention was aimed at the investigation of other 6-alkylpurine derivatives. Recently, cytokinin activity was reported in some 6-(arylalkynyl)-, 6-(arylalkenyl)-, and 6-(arylalkyl)purines.[21] We have recently described the cytostatic activity of 6-(trifluoromethyl)purine riboside.[22] The corticotropin-releasing hormone antagonist activity of some 2,8,9-trisubstituted-6-arylpurines has been also reported.[23]

C-Nucleosites bearing a 2,4-difluoro-5-methylphenyl moiety[24] are recognized by DNA polymerases as analogues of dTMP and, if incorporated into single-stranded

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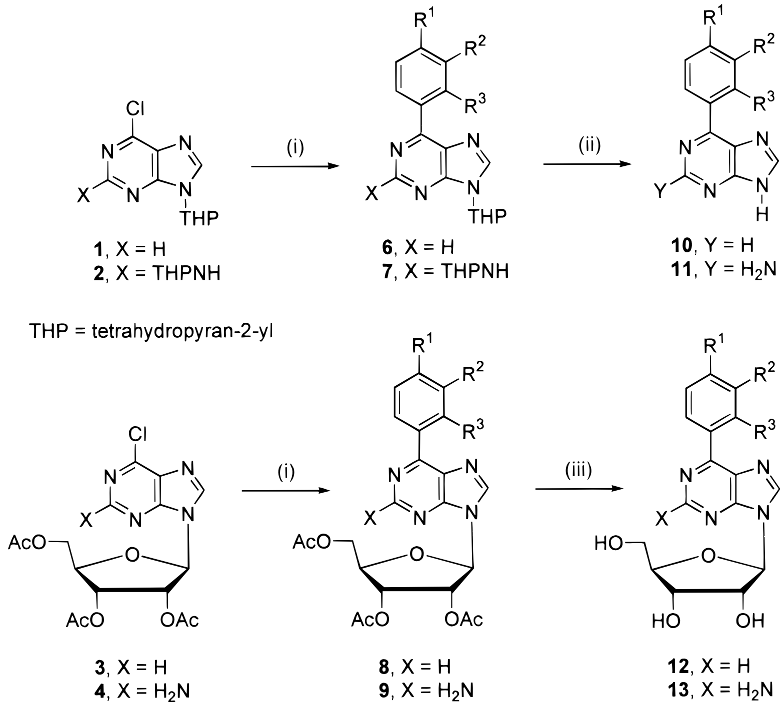
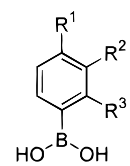
10.1021/jm991167+ CCC: $19.00
Published on Web 04/14/2000

Scheme 1[a]



5 - 13	R^1	R^2	R^3
a	H	H	H
b	F	H	H
c	F	H	F
d	F	F	H

5, 8, 12	R^1	R^2	R^3
e	H	H	CH_3
f	CH_3	H	H
g	CH_3O	H	H
h	H	CH_3O	H
i	EtO	H	H
j	Cl	H	H



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[a] Reagents and conditions: (i) R^1,R^2,R^3PhB(OH)$_2$ (**5**), K_2CO_3, Pd(PPh$_3$)$_4$, toluene; (ii) Dowex 50×8 (H^+), MeOH, H_2O; (iii) NaOMe (0.1 equiv), MeOH.

DNA, encode selectively for adenine in DNA replication. Considering all the above-mentioned findings and surprisingly high biocompatibility of the fluorophenyl group in C-nucleosides, we have focused on the introduction of phenyl and diverse fluorophenyl groups into position 6 of the purine ring. Here we report on the synthesis and in vitro antineoplastic activity of 6-phenylpurine bases and nucleosides against selected transformed cell lines.

Chemistry

With the development of the cross-coupling methodology, many 6-C-substituted purines have been prepared in the past decade. Thus, 6-halopurine derivatives react with arylmagnesium halides,[25] alkyl(aryl)zinc or tin reagents,[26] trialkylaluminum,[27] or alkylcuprates[28] to give the 6-alkylpurine derivatives. Also a reverse approach based on the reaction of purine-6-zinc iodide with aryl or vinyl halides has recently been described.[29] For the synthesis of 6-arylpurines, an alternative approach makes use of radical photochemical reactions of adenine derivatives with aromatic compounds,[30] but this method is very unselective and for substituted benzenes, mixtures of ortho-, meta-, and para-substituted derivatives were obtained.

Recently we have reported a preliminary communication on Suzuki–Miyaura type of cross-coupling of 6-halopurines with aryl- or alkenylboronic acid leading to 6-aryl- and 6-alkenylpurines in good yields.[31] Two different optimized procedures have been elaborated. Anhydrous conditions using K_2CO_3 in toluene were favorable for the coupling of phenylboronic acids bearing an electroneutral or electron-donor substituent, while aqueous conditions using dimethoxyethane and saturated aqueous Na_2CO_3 were successfully used for the coupling reactions of alkenylboronic acids and phenylboronic acids bearing an electron-withdrawing group. Advantages of this reaction compared to the above-mentioned methods are high stability of boronic acids, tolerance toward the presence of diverse functions (including amino and hydroxy groups), low toxicity of boron compounds, easy workup and isolation of the product, and the availability of starting boronic acids (a number of boronic acids, in particular aromatic ones, are commercially available). It is the methodology of choice for the synthesis of a series of substituted 6-phenylpurines and their ribonucleosides.

Tetrahydropyran-2-yl (THP)-protected 6-chloropurine **1**[32] and bis(THP)-protected 2-amino-6-chloropurine **2**,[33] as well as 2′,3′,5′-*O*-triacetyl 6-chloropurine riboside **3**[34] and its 2-amino-6-chloropurine analogue **4**[35] were chosen as suitable starting compounds for the cross-coupling reactions with unsubstituted and fluorinated phenylboronic acids **5a–d**. Due to the lability of the acetyl protection of starting nucleosides **3** and **4**, only the anhydrous conditions could be used. The reactions were performed in toluene in the presence of potassium carbonate (2 equiv) and $Pd(PPh_3)_4$ catalyst (5%) under Ar atmosphere. The corresponding protected 6-phenylpurines **6−9(a−d)** were obtained in good yields of 60−95% after column chromatography. On the other hand, the attempted reactions of 6-chloropurines **1−4** with an extremely electron-poor pentafluorophenylboronic acid were unsuccessful. The THP derivatives **6** and **7** were deprotected by the use[33] of wet Dowex 50×8 (H^+) in methanol to afford the 6-phenylpurine and 2-amino-6-phenylpurine bases **10a−d** and **11a−d**, respectively. The acetyl functions in compounds **8a−d** and **9a−d** were cleaved by methanolysis to afford quantitatively the free nucleosides **12a−d** and **13a−d** that were easily purified by crystallization.

Since in our preliminary cytostatic activity screening the promising cytostatic activity of 6-phenylpurine ribonucleosides **12a−d** had been found (vide infra), further attempts focused on the synthesis of other substituted 6-phenylpurine ribonucleosides bearing diverse types of substituents on the benzene moiety. Thus the 6-chloropurine nucleoside **3** has been submitted in a series of reactions with a variety of substituted phenylboronic acids. In accord with our previous findings,[31] phenylboronic acids bearing electroneutral and electron-donor substituents (methyl, alkoxy, and chloro − compounds **5e−j**) reacted smoothly under analogous conditions giving the corresponding 6-arylpurines **8e−j** in high yields, while phenylboronic acids bearing electron-withdrawing groups (e.g. 3-nitrophenyl, 4-formylphenyl, and 4-acetylphenyl derivatives) did not give cross-coupled products in satisfactory yields. Methanolysis of acetates **8e−j** gave the desired free nucleosides **12e−j**.

In conclusion, the application of the Suzuki−Miyaura reaction of 6-chloropurine derivatives with substituted phenylboronic acids is a facile and effective approach for the synthesis of a series of specifically substituted 6-phenylpurine bases and nucleosides. In comparison with the previously known methods[25−30] using other types of organometallic reagents or photochemistry, this method is more effective and selective, and therefore, further applications in the synthesis of 6-C-substituted purine derivatives may be expected.

All compounds were fully characterized by MS and ^{1}H and ^{19}F NMR; ^{13}C NMR was recorded for at least one example of each class of compounds. UV spectra of the parent unsubstituted 6-phenylpurine bases and ribosides **10a**, **11a**, **12a,** and **13a** were recorded, as well as of the whole series of substituted nucleosides **12a−j** showing expectable substituent effects: bathochromic shifts in compounds bearing electron-donating substituents in the para- or meta-positions of the phenyl ring or hypsochromic shifts in compounds bearing ortho-substituents. All target 6-phenylpurine bases and nucleosides **10−13** were crystallized and characterized by microanalysis. Some oily protected intermediates did not give satisfactory analysis and were used in the deprotection step as TLC homogeneous material. For the assignment of the NMR signals, standard 2D techniques were used. $^{1}H-^{1}H$ spin network was determined by COSY spectra. Carbon connectivities based on one-bond and three-bond $^{1}H-^{13}C$ correlations were established by inverse techniques HMQC and HMBC spectra, respectively. The bis(THP)-protected 2-aminopurines **7** were isolated, characterized, and used as diastereomeric mixtures. They were chromatographically homogeneous, but splitting of some carbon signals was observed in ^{13}C NMR spectra.

Table 1. Cytostatic Activity of 6-Phenylpurine Nucleosides **12** and Their Triacetates **8**

compd	R^1	R^2	R^3	IC_{50} (μmol/L)[a] L1210	HeLa	CCRF-CEM
12a	H	H	H	9.0	2.7	0.7
12b	F	H	H	4.5	2.5	0.75
12c	F	H	F	NA	NA	NA
12d	F	F	H	20.0	2.5	1.4
12e	H	H	Me	NA	NA	NA
12f	Me	H	H	NA	NA	1.5
12g	MeO	H	H	1.5	4.3	0.25
12h	H	MeO	H	NA	NA	4.8
12i	EtO	H	H	2.5	5.0	0.6
12j	Cl	H	H	NA	5.0	0.9
8a	H	H	H	NA	NA	10.5
8b	F	H	H	NA	15.0	4.0
8c	F	H	F	NA	NA	NA
8d	F	F	H	NA	15.0	8.3
8e	H	H	Me	NA	NA	NA
8f	Me	H	H	NA	NA	NA
8g	MeO	H	H	17.0	19.0	4.3
8h	H	MeO	H	NA	NA	NA
8i	EtO	H	H	NA	NA	3.8
8j	Cl	H	H	NA	NA	4.3

[a] NA, not active (inhibition of the cell growth at $c = 10$ μmol/L was lower than 20%).

Cytostatic Activity Evaluation

The title substituted 6-phenylpurine bases and nucleosides **10−13**, as well as some acetyl derivatives **8,** were tested on their in vitro inhibition of the cell growth in the following cell cultures: mouse leukemia L1210 cells (ATCC CCL 219); murine L929 cells (ATCC CCL 1); human cervix carcinoma HeLa S3 cells (ATCC CCL 2.2); and human T-lymphoblastoid CCRF-CEM cell line (ATCC CCL 119). Only substituted 6-phenylpurine ribonucleosides **12** and their triacetates **8** exhibited significant activity in these assays (Table 1), while the bases **10** and **11**, as well as the 2-amino-6-phenylpurine ribonucleosides **13,** were entirely inactive.

The results (Table 1) show that the 6-phenylpurine ribonucleosides **12** possess powerful cytostatic potency against T-lymphoblastoids (CCRF-CEM: IC_{50} values from 0.25 to 8.3 μmol/L) while HeLa S3 cells are less sensitive (IC_{50} values from 2.5 to 19 μmol/L). Markedly reduced cytostatic activity against L1210 cells and marginal effects toward L929 cell line (data not shown) might reflect a difference in the transport mechanism of mentioned compounds into the different cell lines and/or a putative process of the metabolic activation.

The structure−activity relationship of the series of compounds shows that the most active compounds are 6-phenylpurine nucleoside **12a** and its congeners bearing a functionality in position 4 of the benzene ring (compounds **12b**, **12g**, **12i**, and **12j**). Introduction of a substituent into position 3 of the benzene moiety causes a substantial decrease of activity (compounds **12d** and **12h**), while the presence of a substituent in position 2 leads to inactive compounds (**12c** and **12e**). Also the presence of an amino function in position 2 of the purine ring causes loss of activity. The acetyl derivatives **8** of active compounds are considerably less active than the parent free nucleosides **12**.

In conclusion, a novel type of antineoplastic compounds, substituted 6-phenylpurine ribonucleosides, has been discovered. In our view, the cytostatic activity of this class of compounds cannot be deduced from any known cytostatic nucleoside or purine derivatives, and therefore, it might be considered a new structural lead in the search for antitumor compounds.

Experimental Section

Unless otherwise stated, solvents were evaporated at 40 °C/2 kPa and compounds were dried at 60 °C/2 kPa over P_2O_5. Melting points were determined on a Kofler block and are uncorrected. NMR spectra were measured on Bruker AMX-3 400 (400 MHz for ^{1}H, 100.6 MHz for ^{13}C and 376.5 MHz for ^{19}F nuclei), Bruker DRX 500 (500 MHz for ^{1}H, 125.7 MHz for ^{13}C and 470.59 MHz for ^{19}F) and Varian Gemini 300HC (300.075 MHz for ^{1}H and 75.462 MHz for ^{13}C). TMS was used as internal standard for the ^{1}H and ^{13}C NMR spectra; $CFCl_3$ was an internal standard for ^{19}F spectra; values are given in δ (ppm) and J values are in Hz. Mass spectra were measured on a ZAB-EQ (VG Analytical) spectrometer using FAB (ionization by Xe, accelerating voltage 8 kV, glycerol matrix) or EI (electron energy 70 eV) techniques. Toluene was degassed in vacuo and stored over molecular sieves under Ar. Substituted phenylboronic acids **5** were supplied by Aldrich.

Suzuki Coupling of the 6-Chloropurines and Substituted Phenylboronic Acids: General Procedure. Toluene (10 mL) was added to an argon-purged flask containing a 6-chloropurine **1**−**4** (1 mmol), K_2CO_3 (200 mg, 1.5 mmol), substituted phenylboronic acid **5** (1.5 mmol) and $Pd(PPh_3)_4$ (59 mg, 0.05 mmol) and the mixture was stirred under argon at 100 °C for 8 h. After cooling to ambient temperature the mixture was evaporated in vacuo and the residue was chromatographed on a silica gel column (50 g, ethyl acetate−light petroleum 1:2 to 9:1). Evaporation and drying of the product containing fractions afforded the 6-phenylpurines **6**−**9** as foams or amorphous solids.

6-Phenyl-9-(tetrahydropyran-2-yl)purine (6a). Colorless amorphous solid, yield 95%. FAB MS m/z (rel. %): 281 (40) [M + H], 197 (100) [M + H − THP], 85 (14) [THP]. ^{1}H NMR (400 MHz, $CDCl_3$): 1.6−1.9 and 2.0−2.2 (2 × m, 6 H, CH_2); 3.82 (dt, 1 H, J = 2.5, 11.6, H-5′a); 4.21 (m, 1 H, H-5′b); 5.86 (dd, 1 H, J = 10.4 and 2.7, H-1′); 7.5−7.6 (m, 3 H, H−Ar); 8.34 (s, 1 H, H-8); 8.77−8.80 (m, 2 H, H−Ar); 9.03 (s, 1 H, H-2). Anal. ($C_{16}H_{16}N_4O$) C, H, N.

6-(4-Fluorophenyl)-9-(tetrahydropyran-2-yl)purine (6b). Colorless amorphous solid, yield 84%. FAB MS m/z (rel. %): 299 (7) [M + H], 215 (100) [M + H − THP]. ^{1}H NMR (400 MHz, $CDCl_3$): 1.6−1.9 and 2.0−2.2 (2 × m, 6 H, CH_2); 3.81 (dt, 1 H, J = 2.2 and 11.5, H-5′a); 4.20 (brd, 1 H, J = 11.5, H-5′b); 5.84 (dd, 1 H, J = 10.3 and 2.3, H-1′); 7.22 (t, 2 H, J = 8.7, H−o-Ar); 8.31 (s, 1 H, H-8); 8.84 (dd, 2 H, J = 8.7 and 5.7, H−m-Ar); 8.99 (s, 1 H, H-2). ^{13}C NMR (100 MHz, $CDCl_3$): 22.78, 24.87 and 31.86 (CH_2); 68.87 (CH_2-5′); 82.02 (CH-1′); 115.67 (d, J = 21.4, m-CH−FPh); 130.84 (C-5); 131.73 (i-C−FPh); 132.01 (d, J = 8.5, o-CH−FPh); 142.04 (C-8); 152.36 (C-2); 151.71 and 153.71 (C-4 and C-6); 164.64 (d, ^1J(C,F) = 252.0, CF). ^{19}F NMR (376.5 MHz, $CDCl_3$): −109.56 (s, F−Ph). Anal. ($C_{16}H_{15}FN_4O$) C, H, N.

6-(2,4-Difluorophenyl)-9-(tetrahydropyran-2-yl)purine (6c). Colorless amorphous solid, yield 66%. FAB MS m/z (rel. %): 317 (19) [M + H], 233 (100) [M + H − THP], 85 (30) [THP]. ^{1}H NMR (400 MHz, $CDCl_3$): 1.60−1.85 and 2.0−2.2 (2 × m, 6 H, CH_2); 3.75−3.85 (m, 1 H, H-5′a); 4.16−4.23 (m, 1 H, H-5′b); 5.76−5.88 (m, 1 H, H-1′); 6.97−7.09 (m, 2 H, H−Ar); 8.04 (q, 1 H, J = 6.7, H−Ar); 8.34 (s, 1 H, H-8); 9.07 (s, 1 H, H-2). ^{19}F NMR (376.5 MHz, $CDCl_3$): −106.90 and −108.35 (2 × m, F_2Ph).

6-(3,4-Difluorophenyl)-9-(tetrahydropyran-2-yl)purine (6d). Colorless amorphous solid, yield 70%. FAB MS m/z (rel. %): 317 (24) [M + H], 233 (100) [M + H − THP]. ^{1}H NMR (400 MHz, $CDCl_3$): 1.6−1.9 and 2.0−2.2 (2 × m, 6 H, CH_2); 3.82 (dt, 1 H, J = 2.6 and 11.6, H-5′a); 4.21 (m, 1 H, H-5′b); 5.85 (dd, 1 H, J = 10.3 and 2.5, H-1′); 7.33 (m, 1 H, H−Ar); 8.34 (s, 1 H, H-8); 8.66−8.78 (m, 2 H, H−Ar); 8.99 (s, 1 H, H-2). ^{19}F NMR (376.5 MHz, $CDCl_3$): −134.26 (dd, J_1 = J_2 = 9.4) and −137.52 (pent, J = 10.8, F_2Ph). Anal. ($C_{16}H_{14}F_2N_4O$) C, H, N.

6-Phenyl-9-(tetrahydropyran-2-yl)-2-[(tetrahydropyran-2-yl)amino]purine (7a). Yellow amorphous solid, yield 95%. FAB MS m/z (rel. %) 380 (25) [M + H], 212 (100) [M + 2H − 2THP]. ^{1}H NMR (400 MHz, $CDCl_3$): 1.5−2.1 (m, 12 H, CH_2); 3.66−3.78 (m, 2 H, H-5′ and H-5"a); 4.05 (d, 1 H, J = 12.8) and 4.15 (d, 1 H, J = 12.1, H-5′ and H-5"b); 5.53 (t, 1 H, J = 9.2) and 5.64−5.82 (m, 2 H, H-1′, H-1" and NH); 7.46−7.53 (m, 3 H, H−Ar); 8.03 (s, 1 H, H-8); 8.66−8.70 (m, 2 H, H−Ar). ^{13}C NMR (100 MHz, $CDCl_3$): 22.97, 23.03, 24.96, 25.35, 31.33, 31.61 and 31.78 (CH_2); 66.49, 66.57 and 68.60 (CH_2-5′ and CH_2-5″); 80.32, 80.47, 81.18 and 81.62 (CH-1′ and CH-1″); 125.96 and 126.03 (C-5); 128.73, 129.61 and 130.55 (CH−Ph); 136.04 and 136.10 (i-C−Ph); 139.27 and 139.41 (C-8); 153.51, 153.64, 155.54 and 155.67 (C-4 and C-6); 157.75 and 157.84 (C-2).

6-(4-Fluorophenyl)-9-(tetrahydropyran-2-yl)-2-[(tetrahydropyran-2-yl)amino]purine (7b). Colorless amorphous solid, yield 81%. FAB MS m/z (rel. %): 398 (33) [M + H], 230 (100) [M + 2H − 2THP]. ^{1}H NMR (400 MHz, $CDCl_3$): 1.5−2.2 (m, 12 H, CH_2); 3.68−3.78 (m, 2 H, H-5′ and H-5"a); 4.04 (d, 1 H, J = 12.2) and 4.16 (d, 1 H, J = 11.8, H-5′ and H-5"b); 5.50 (t, 1 H) and 5.66 (t, 1 H, J = 10.1, H-1′ and H-1"); 7.18 (t, 2 H, J = 8.6, H−o-Ar); 8.00 (s, 1 H, H-8); 8.71−8.75 (m, 2 H, H−m-Ar). ^{13}C NMR (100 MHz, $CDCl_3$): 22.97, 23.02, 24.95, 25.33, 31.35, 31.61 and 31.78 (CH_2); 66.52, 66.58 and 68.62 (CH_2-5′ and CH_2-5″); 80.31, 80.45, 81.22 and 81.62 (CH-1′ and CH-1″); 115.36 (d, J = 21.3, m-CH−FPh); 125.69 and 125.76 (C-5); 131.79 (d, J = 8.3, o-CH−FPh); 132.26 (i-C−FPh); 139.31 and 139.44 (C-8); 153.54, 153.67, 154.26 and 154.38 (C-4 and C-6); 157.70 and 157.78 (C-2); 164.42 (d, ^1J(CF) = 251.0, CF). ^{19}F NMR (376.5 MHz, $CDCl_3$): −110.37 (s, F−Ph). Anal. ($C_{21}H_{24}FN_5O_2$) C, H; N: calcd, 17.62; found, 17.12.

6-(2,4-Difluorophenyl)-9-(tetrahydropyran-2-yl)-2-[(tetrahydropyran-2-yl)amino]purine (7c). Colorless amorphous solid, yield 60%. FAB MS m/z (rel. %): 416 (76) [M + H], 332 (100) [M + H − THP], 248 (66) [M + 2H − 2THP]. ^{1}H NMR (400 MHz, $CDCl_3$): 1.5−2.2 (m, 12 H, CH_2); 3.65−3.82 (m, 2 H, H-5′and H-5"a); 4.03 (d, 1 H, J = 12.0) and 4.17 (d, 1 H, J = 11.8, H-5′ and H-5"b); 5.43−5.50 (m, 1 H) and 5.65−5.77 (m, 2 H, NH, H-1′ and H-1"); 6.93−7.04 (m, 2 H, H−Ar); 7.91−8.00 (m, 1 H, H−Ar); 8.01 (s, 1 H, H-8). ^{19}F NMR (376.5 MHz, $CDCl_3$): −107.83 (brs, F−Ph). Anal. ($C_{21}H_{23}F_2N_5O_2$) C, H, N.

6-(3,4-Difluorophenyl)-9-(tetrahydropyran-2-yl)-2-[(tetrahydropyran-2-yl)amino]purine (7d). Yellow amorphous solid, yield 60%. FAB MS m/z (rel. %): 416 (88) [M + H], 332 (100) [M + H − THP], 248 (93) [M + 2H − 2THP]. ^{1}H NMR (400 MHz, $CDCl_3$): 1.5−2.2 (m, 12 H, CH_2); 3.65−3.80 (m, 2 H, H-5′and H-5"a); 4.04 (d, 1 H, J = 11.5) and 4.15 (d, 1 H, J = 10.9, H-5′ and H-5"b); 5.50 (brt, 1 H, J = 9.6) and 5.62−5.77 (m, 2 H, NH, H-1′ and H-1"); 7.23−7.31 (m, 1 H, H−Ar); 8.03 (s, 1 H, H-8); 8.55−8.62 (m, 2 H, H−Ar). ^{19}F NMR (376.5 MHz, $CDCl_3$): −134.93 and −137.95 (2 × brs, F_2Ph).

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-6-phenylpurine (8a). Colorless amorphous solid, yield 79%. FAB MS m/z (rel. %): 455 (22) [M + H], 197 (100) [M + H − AcRf]. ^{1}H NMR (400 MHz, $CDCl_3$): 2.09, 2.13 and 2.16 (3 × s, 3 × 3 H, CH_3); 4.37−4.50 (m, 3 H, H-4′ and 2 × H-5′); 5.71 (dd, 1 H, J = 3.6 and 5.4, H-3′); 6.02 (t, 1 H, J = 5.4, H-2′); 6.30 (d, 1 H, J = 5.3, H-1′); 7.52−7.58 (m, 3 H, H−Ph); 8.28 (s, 1 H, H-8); 8.76 (dd, 2 H, J = 1.6 and 8.0, H−Ph); 9.03 (s, 1 H, H-2). Anal. ($C_{22}H_{22}N_4O_7$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-6-(4-fluorophenyl)purine (8b). Colorless amorphous solid, yield 87%. FAB MS m/z (rel. %): 473 (8) [M + H], 215 (100) [M + H − AcRf]. ^{1}H NMR (400 MHz, $CDCl_3$): 2.07, 2.12 and 2.15 (3 × s, 3 × 3 H, CH_3); 4.35−4.47 (m, 3 H, H-4 and 2 × H-5); 5.69 (dd, 1 H, J = 4.1 and 5.2, H-3′); 6.00 (t, 1 H, J = 5.2, H-2′); 6.27 (d, 1 H, J = 5.1, H-1′); 7.22 (t, 2 H, J = 8.4, H−o-Ar); 8.26 (s, 1 H, H-8); 8.82 (m, 2 H, H−m-Ar); 8.98 (s 1 H, H-2). ^{13}C NMR (100 MHz, $CDCl_3$): 21.05, 21.19 and 21.42 (CH_3); 63.75 (CH_2-5′); 71.37 (CH-3′); 73.82 (CH-2′); 81.13 (CH-4′); 87.17 (CH-1′); 116.46 (d, J = 21.6, *m*-CH−FPh); 132.07 (*i*-C−FPh); 132.29 (C-5); 132.81 (d, J = 8.3, *o*-CH−FPh); 143.23 (C-8); 152.76 (C-4); 153.34 (C-2); 154.93 (C-6), 165.48 (d, ^1J(C,F) = 251.0, CF); 170.03, 170.24 and 170.96 (3 × CO). ^{19}F NMR (376.5 MHz, $CDCl_3$): −109.12 (s, F−Ph). Anal. ($C_{22}H_{21}FN_4O_7$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-6-(2,4-difluorophenyl)purine (8c). Colorless amorphous solid, yield 65%. FAB MS *m*/*z* (rel. %): 491 (19) [M + H], 233 (100) [M + H − AcRf]. ^{1}H NMR (400 MHz, $CDCl_3$): 2.11, 2.13 and 2.17 (3 × s, 3 × 3 H, CH_3); 4.38−4.50 (m, 3 H, H-4 and 2 × H-5); 5.71 (dd, 1 H, J = 4.6 and 5.4, H-3′); 6.03 (t, 1 H, J = 5.4, H-2′); 6.30 (d, 1 H, J = 5.3, H-1′); 7.00−7.12 (m, 2 H, H−Ar); 8.03 (m, 1 H, H−Ar); 8.28 (s, 1 H, H-8); 9.09 (s, 1 H, H-2). ^{19}F NMR (376.5 MHz, $CDCl_3$): −106.45 (t, J = 7.3, FPh); −108.18 (q, J = 8.8, FPh). Anal. ($C_{22}H_{20}F_2N_4O_7$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-6-(3,4-difluorophenyl)purine (8d). Colorless amorphous solid, yield 81%. FAB MS *m*/*z* (rel. %): 491 (100) [M + H], 233 (66) [M + H − AcRf]. ^{1}H NMR (400 MHz, $CDCl_3$): 2.09, 2.14 and 2.17 (3 × s, 9 H, CH_3); 4.38−4.50 (m, 3 H, H-4 and 2 × H-5); 5.70 (m, 1 H, H-3′); 6.00 (m, 1 H, H-2′); 6.28 (d, 1 H, J = 5.2, H-1′); 7.29−7.38 (m, 1 H, H−Ar); 8.28 (s, 1 H, H-8); 8.64−8.77 (m, 2 H, H−Ar); 9.01 (s, 1 H, H-2). ^{19}F NMR (376.5 MHz, $CDCl_3$): −133.76 and −137.32 (2 × m, F_2Ph). Anal. ($C_{22}H_{20}F_2N_4O_7$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-6-(2-tolyl)purine (8e). Colorless amorphous solid, yield 89%. FAB MS *m*/*z* (rel. %): 469 (18) [M + H], 211 (100) [M + H − AcRf]. ^{1}H NMR (400 MHz, $CDCl_3$): 2.10, 2.11 and 2.15 (3 × s, 3 × 3 H, CH_3); 2.43 (s, 3 H, CH_3Ph); 4.37−4.49 (m, 3 H, H-4′ and 2 × H-5′); 5.72 (dd, 1 H, J = 5.0 and 5.3, H-3′); 6.02 (t, 1 H, J = 5.3, H-2′); 6.27 (d, 1 H, J = 5.1, H-1′); 7.7.3−7.4 (m, 3 H, H−Ar); 7.66−7.69 (m, 1 H, H−Ar); 8.21 (s, 1 H, H-8); 9.05 (s 1 H, H-2). ^{13}C NMR (100 MHz, $CDCl_3$): 21.05−21.27 (m, 4 × CH_3); 63.59 (C-5′); 71.20 (C-3′); 73.71 (C-2′); 80.98 (C-4′); 87.22 (C-1′); 126.34, 130.38, 131.34, 131.73 (CH-arom); 135.25, 137.75 (C-arom); ∼143 (very weak, C-8); 151.93 (C-4); 153.07 (C-2); 160.17 (C-6), 169.89−170.80 (m, 3 × CO). Anal. ($C_{23}H_{24}N_4O_7$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-6-(4-tolyl)purine (8f). Colorless amorphous solid, yield 79%. FAB MS *m*/*z* (rel. %): 469 (22) [M + H], 211 (100) [M + H − AcRf]. ^{1}H NMR (400 MHz, $CDCl_3$): 2.07, 2.12 and 2.15 (3 × s, 3 × 3 H, CH_3); 2.44 (s, 3 H, CH_3Ph); 4.36−4.48 (m, 3 H, H-4′ and 2 × H-5′); 5.70 (dd, 1 H, J = 4.5 and 5.4, H-3′); 6.00 (t, 1 H, J = 5.4, H-2′); 6.28 (d, 1 H, J = 5.3, H-1′); 7.36 (d, 2 H, J = 8.1, H−Ph); 8.24 (s, 1 H, H-8); 8.67 (d, 2 H, J = 8.1, H−Ph); 8.99 (s, 1 H, H-2). Anal. ($C_{23}H_{24}N_4O_7$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-6-(4-methoxyphenyl)purine (8g). Colorless amorphous solid, yield 84%. FAB MS *m*/*z* (rel. %): 485 (24) [M + H], 227 (100) [M + H − AcRf]. ^{1}H NMR (400 MHz, $CDCl_3$): 2.07, 2.12 and 2.14 (3 × s, 3 × 3 H, CH_3); 3.88 (s, 3 H, OCH_3); 4.36−4.48 (m, 3 H, H-4′ and 2 × H-5′); 5.69 (dd, 1 H, J = 4.5 and 5.4, H-3′); 5.99 (t, 1 H, J = 5.4, H-2′); 6.28 (d, 1 H, J = 5.4, H-1′); 7.06 (d, 2 H, J = 9.0, H−Ph); 8.22 (s, 1 H, H-8); 8.79 (d, 2 H, J = 9.0, H−Ph); 8.95 (s, 1 H, H-2). Anal. ($C_{23}H_{24}N_4O_8$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-6-(3-methoxyphenyl)purine (8h). Colorless amorphous solid, yield 74%. FAB MS *m*/*z* (rel. %): 485 (13) [M + H], 227 (100) [M + H − AcRf]. ^{1}H NMR (400 MHz, $CDCl_3$): 2.12, 2.17 and 2.19 (3 × s, 3 × 3 H, CH_3); 3.96 (s, 3 H, CH_3O); 4.41−4.52 (m, 3 H, H-4′ and 2 × H-5′); 5.73 (dd, 1 H, J = 4.5 and 5.4, H-3′); 6.04 (t, 1 H, J = 5.4, H-2′); 6.33 (d, 1 H, J = 5.3, H-1′); 7.12 (dd, 1 H, J = 8.0, 2.6, H−Ph); 7.50 (t, 1 H, J = 8.0, H−Ph); 8.30 (s, 1 H, H-8); 8.36 (dd, 1 H, J = 1.9, 2.4, H−Ph); 8.44 (d, 1 H, J = 7.8, H−Ph); 9.05 (s 1 H, H-2). Anal. ($C_{23}H_{24}N_4O_8$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-6-(4-ethoxyphenyl)purine (8i). Colorless amorphous solid, yield 80%. FAB MS *m*/*z* (rel. %): 499 (30) [M + H], 241 (100) [M + H − AcRf]. ^{1}H NMR (500 MHz, $CDCl_3$): 1.46 (t, 3 H, J = 6.9, CH_3CH_2); 2.09, 2.14 and 2.16 (3 × s, 3 × 3 H, CH_3); 4.14 (q, 2 H, J = 6.9, CH_2CH_3); 4.38−4.49 (m, 3 H, H-4′ and 2 × H-5′); 5.71 (dd, 1 H, J = 4.3 and 5.4, H-3′); 6.01 (t, 1 H, J = 5.4, H-2′); 6.29 (d, 1 H, J = 5.3, H-1′); 7.06 (d, 2 H, J = 8.8, H−Ph); 8.24 (s, 1 H, H-8); 8.78 (d, 2 H, J = 8.8, H−Ph); 8.96 (s, 1 H, H-2). Anal. ($C_{24}H_{26}N_4O_8$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-6-(4-chlorophenyl)purine (8j). Colorless amorphous solid, yield 65%. FAB MS *m*/*z* (rel. %): 489 (19) [M + H], 231 (100) [M + H − AcRf]. ^{1}H NMR (500 MHz, $CDCl_3$): 2.09, 2.14 and 2.17 (3 × s, 3 × 3 H, CH_3); 4.39−4.51 (m, 3 H, H-4′ and 2 × H-5′); 5.71 (dd, 1 H, J = 4.7 and 5.4, H-3′); 6.01 (t, 1 H, J = 5.4, H-2′); 6.30 (d, 1 H, J = 5.3, H-1′); 7.53 (d, 2 H, J = 8.6, H−Ph); 8.29 (s, 1 H, H-8); 8.77 (d, 2 H, J = 8.6, H−Ph); 9.02 (s, 1 H, H-2). Anal. ($C_{22}H_{21}ClN_4O_7$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-2-amino-6-phenylpurine (9a). Yellowish amorphous solid, yield 83%. FAB MS *m*/*z* (rel. %): 470 (30) [M + H], 212 (100) [M + H − AcRf]. ^{1}H NMR (400 MHz, $CDCl_3$): 2.09, 2.10 and 2.15 (3 × s, 3 × 3 H, CH_3); 4.35−4.50 (m, 3 H, H-4 and 2 × H-5); 5.13 (s, 2 H, NH_2); 5.84 (dd, 1 H, J = 4.5 and 5.3, H-3′); 6.04 (dd, 1 H, J = 4.9 and 5.3, H-2′); 6.08 (d, 1 H, J = 4.9, H-1′); 7.49−7.54 (m, 3 H, H−Ph); 7.90 (s, 1 H, H-8); 8.60−8.63 (m, 2 H, H−Ph). Anal. ($C_{22}H_{23}N_5O_7$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-2-amino-6-(4-fluorophenyl)purine (9b). Colorless amorphous solid, yield 80%. FAB MS *m*/*z* (rel. %): 488 (14) [M + H], 230 (100) [M + H − AcRf]. ^{1}H NMR (400 MHz, $CDCl_3$): 2.08, 2.09 and 2.14 (3 × s, 3 × 3 H, CH_3); 4.35−4.50 (m, 3 H, H-4 and 2 × H-5); 5.10 (s, 2 H, NH_2); 5.82 (t, 1 H, J = 4.8, H-3′); 6.02 (t, 1 H, J = 4.9, H-2′); 6.07 (d, 1 H, J = 4.8, H-1′); 7.20 (m, 2 H, H−o-Ar); 7.89 (s, 1 H, H-8). 8.69 (m, 2 H, H−m-Ar). ^{13}C NMR (100 MHz, $CDCl_3$): 21.41, 20.52 and 20.68 (CH_3); 63.04 (CH_2-5′); 70.61 (CH-3′); 72.79 (CH-2′); 79.89 (CH-4′); 86.32 (CH-1′); 115.49 (d, J = 21.1, *m*-CH−FPh); 125.80 (C-5); 131.78 (d, J = 8.7, *o*-CH−FPh); 139.91 (C-8); 153.80 (C-4); 155.21 (C-6); 159.52 (C-2), 164.55 (d, ^1J(C,F) = 251.6, CF); 169.35, 169.56 and 170.47 (3 × CO). ^{19}F NMR (376.5 MHz, $CDCl_3$): −109.81 (s, F−Ph). Anal. ($C_{22}H_{22}FN_5O_7$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-2-amino-6-(2,4-difluorophenyl)purine (9c). Colorless amorphous solid, yield 75%. FAB MS *m*/*z* (rel. %): 506 (27) [M + H], 248 (100) [M + H − AcRf]. ^{1}H NMR (400 MHz, $CDCl_3$): 2.08, 2.11 and 2.16 (3 × s, 3 × 3 H, CH_3); 4.35−4.50 (m, 3 H, H-4 and 2 × H-5); 5.20 (s, 2 H, NH_2); 5.83 (dd, 1 H, J = 4.3 and 5.2, H-3′); 6.04 (dd, 1 H, J = 4.9 and 5.2, H-2′); 6.07 (d, 1 H, J = 4.9, H-1′); 6.95−7.06 (m, 2 H, H−Ar); 7.85−7.91 (m, 1 H, H−Ar); 7.89 (s, 1 H, H-8). ^{19}F NMR (376.5 MHz, $CDCl_3$): −108.20 and −107.22 (2 × m, F_2Ph). Anal. ($C_{22}H_{21}F_2N_5O_7$) C, H, N.

9-(2,3,5-Tri-*O*-acetyl-*β*-D-ribofuranosyl)-2-amino-6-(3,4-difluorophenyl)purine (9d). Colorless amorphous solid, yield 81%. FAB MS *m*/*z* (rel. %): 506 (35) [M + H], 248 (100) [M + H − AcRf]. ^{1}H NMR (400 MHz, $CDCl_3$): 2.10, 2.11 and 2.16 (3 × s, 3 × 3 H, CH_3); 4.35−4.50 (m, 3 H, H-4 and 2 × H-5); 5.12 (s, 2 H, NH_2); 5.83 (t, 1 H, J = 5.0, H-3′); 6.03 (t, 1 H, J = 5.0, H-2′); 6.08 (d, 1 H, J = 4.9, H-1′); 7.25−7.33 (m, 1 H, H−Ar); 7.91 (s, 1 H, H-8); 8.52−8.62 (m, 2 H, H−Ar). ^{19}F NMR (376.5 MHz, $CDCl_3$): −137.75 and −134.48 (2 × m, F_2-Ph). Anal. ($C_{22}H_{21}F_2N_5O_7$) C, H, N.

Cleavage of the THP-Protected Purines 6 and 7: General Procedure. A mixture of a THP-protected base **6** or **7** (0.6−0.8 mmol), Dowex 50×8 (H^+) (ca. 300 mg), methanol (10 mL) and water (1 mL) was refluxed for 1 h, then filtered while hot and the resin was washed with saturated methanolic ammonia (5 mL) followed by methanol (20 mL). The combined filtrates were evaporated and the residue was codistilled with toluene. Crystallization of the residue from methanol/toluene with an addition of heptane afforded the free bases **10** or **11**.

6-Phenylpurine (10a). Colorless crystals, yield 92%, mp 280−282 °C (lit.[36] 243 °C). EI MS *m*/*z* (rel. %): 196 (67) [M], 169 (54) [M − HCN], 141 (20) [M − 2 HCN], 41 (100). ^{1}H NMR (400 MHz, DMSO-d_6): 7.54−7.62 (m, 3 H, H−Ar); 8.64 (s, 1 H, H-8); 8.83 (brm, 2 H, H−Ar); 8.96 (s, 1 H, H-2). UV (λ_{max}

(ϵ)) methanol: 289 (12900); water: pH 7, 288 (8600); pH 2, 298 (42100); pH 11, 298 (34700). Anal. ($C_{11}H_8N_4$) C, H, N.

6-(4-Fluorophenyl)purine (10b). Colorless crystals, yield 83%, mp 299−302 °C. EI MS *m/z* (rel. %): 214 (100) [M], 187 (46) [M − HCN]. ^{1}H NMR (400 MHz, $CDCl_3$): 7.44 (t, 2 H, *J* = 8.9, H-*o*-Ar); 8.67 (s, 1 H, H-8); 8.90−9.00 (brm, 2 H, H-*m*-Ar); 8.96 (s, 1 H, H-2); 13.65 (brs, 1 H, NH). ^{13}C NMR (100 MHz, $CDCl_3$): 115.49 and 115.70 (CH-arom); 131.60 and 131.68 (CH-arom); 132.13 (C-5); 144.95 (C-8); ~151 (very weak, C-4); 151.77 (C-2); ~154 (very weak, C-6); 163.71 (d, J(C,F) = 247.4, CF). ^{19}F NMR (376.5 MHz, $CDCl_3$): −109.24 (s, FPh). Anal. ($C_{11}H_7FN_4$) C, H. N.

6-(2,4-Difluorophenyl)purine (10c). Yellowish crystals, yield 81%, mp 304−307 °C. EI MS *m/z* (rel. %): 232 (100) [M], 205 (58) [M − HCN]. ^{1}H NMR (400 MHz, DMSO-d_6): 7.32 (m, 1 H, H−Ar); 7.48 (m, 1 H, H−Ar); 8.04 (br, 1 H, H−Ar); 8.65 (s, 1 H, H-8); 9.00 (s, 1 H, H-2); 13.56 (brs, 1 H, NH). ^{19}F NMR (376.5 MHz, DMSO-d_6): −106.71 and −107.35 (2 × brs, F_2-Ph). Anal. ($C_{11}H_6F_2N_4$) C, H, N.

6-(3,4-Difluorophenyl)purine (10d). Colorless crystals, yield 85%, mp 283−285 °C. EI MS *m/z* (rel. %): 232 (100) [M], 205 (52) [M − HCN]. ^{1}H NMR (400 MHz, DMSO-d_6): 7.67 (m, 1 H, H−Ar); 8.69 (s, 1 H, H-8); 8.73−8.77 (brm, 1 H, H−Ar); 8.80−8.87 (m, 1 H, H−Ar); 8.96 (s, 1 H, H-2). ^{19}F NMR (376.5 MHz, DMSO-d_6): −134.62 and −137.36 (2 × m, F_2Ph). Anal. ($C_{11}H_6F_2N_4$) C, H, N.

2-Amino-6-phenylpurine (11a). Yellowish crystals, yield 88%, mp 111−114 °C (loss of H_2O), 249−252 °C (lit.[37] 257−259 °C). EI MS *m/z* (rel. %): 211 (30) [M], 91 (90), 43 (100). ^{1}H NMR (400 MHz, DMSO-d_6): 6.41 (brs, 2 H, NH_2); 7.50−59 (m, 3 H, H−Ar); 8.14 (s, 1 H, H-8); 8.68−8.72 (brm, 2 H, H−Ar). UV (λ_{max} (ϵ)) methanol: 334 (7900), 260 sh (11500), 243 (14300); water: pH 7, 327 (8400), 258 sh (10800), 244 (12600); pH 2, 340 (10200), 259 (9100); pH 11, 331 (8100). Anal. ($C_{11}H_9N_5$·1/2H_2O) C, H, N.

2-Amino-6-(4-fluorophenyl)purine (11b). Yellowish crystals, yield 78%, mp 261−264 °C (H_2O). EI MS *m/z* (rel. %): 229 (100) [M], 202 (6) [M − HCN], 149 (37). ^{1}H NMR (400 MHz, DMSO-d_6): 6.40 (brs, 2 H, NH_2); 7.38 (dt, 1 H, *J* = 8.9 and 2.0, H-*o*-Ar); 8.14 (s, 1 H, H-8); 8.82 (m, 1 H, H−Ar); 12.70 (brs, 1 H, NH). ^{13}C NMR (100 MHz, DMSO-d_6): 115.09 and 115.30 (CH-arom); 132.40 (C−Ph); 131.29 and 131.37 (CH-arom); 132.64 (C-5); 140.85 (C-8); 151.63 (C-4); 155.63 (C-6); 160.05 (C-2); 163.42 (d, *J* = 246.6, CF). ^{19}F NMR (376.5 MHz, DMSO-d_6): −110.10 (s, FPh). Anal. ($C_{11}H_8FN_5$·H_2O) C, H, N.

2-Amino-6-(2,4-difluorophenyl)purine (11c). Yellowish crystals, yield 78%, mp 252−254 °C. EI MS *m/z* (rel. %): 247 (100) [M], 228 (29) [M − F], 220 (10) [M − HCN]. ^{1}H NMR (400 MHz, DMSO-d_6): 6.49 (brs, 2 H, NH_2); 7.25 (dt, 1 H, *J* = 8.5 and 2.3, H−Ar); 7.40 (dt, 1 H, *J* = 10.4 and 2.4, H−Ar); 7.89 (q, 1 H, *J* = 8.5, H−Ar); 8.12 (s, 1 H, H-8). ^{19}F NMR (376.5 MHz, DMSO-d_6): −107.43 (q, *J* = 9.0) and −107.60 (pent, *J* = 7.9, F_2Ph). Anal. ($C_{11}H_7F_2N_5$·1/2H_2O) C, H, N.

2-Amino-6-(3,4-difluorophenyl)purine (11d). Yellowish crystals, yield 80%, mp 295−298 °C. EI MS *m/z* (rel. %): 247 (100) [M], 220 (5) [M − HCN]. ^{1}H NMR (400 MHz, DMSO-d_6): 6.37 (s, 2 H, NH_2); 7.62 (m, 1 H, H−Ar); 8.14 (s, 1 H, H-8); 8.65−8.69 (brm, 1 H, H−Ar); 8.76 (ddd, 1 H, *J* = 1.9, 8.2 and 10.2, H−Ar). ^{19}F NMR (376.5 MHz, DMSO-d_6): −135.70 and −138.01 (2 × m, F_2Ph). Anal. ($C_{11}H_7F_2N_5$) C, H, N.

Deacetylation of the Protected Nucleosides 8 and 9: General Procedure. A 1 M solution of NaOMe (200 μL, 0.2 mmol) was added to the solution of the protected nucleoside **8** or **9** (0.5−0.8 mmol) in MeOH (20 mL) and the mixture was stirred at ambient temperature overnight. The crystals (if formed) were filtered off. Then the solution was neutralized by addition of Dowex 50×8 (H^+) (ca. 100 mg) and filtered. The ion-exchanger was washed with saturated methanolic ammonia (5 mL) followed by methanol (20 mL) and the combined filtrates were evaporated to dryness. The collected crystals and residue was recrystallized from EtOH/toluene to give the nucleosides **12** or **13**.

6-Phenyl-9-(β-D-ribofuranosyl)purine (12a). Colorless crystals, yield 66%, mp 228−230 °C; $[\alpha]_D$ −56.1 (*c* 0.5 DMF). FAB MS *m/z* (rel. %): 329 (35) [M + H], 197 (75) [M + H − Rf]. ^{1}H NMR (400 MHz, DMSO-d_6): 3.31 (s, 3 H, CH_3); 3.59−3.65 and 3.70−3.76 (2 × m, 2 H, CH_2-5′); 4.02 (m, 1 H, H-4′); 4.23 (m, 1 H, H-3′); 4.67 (ddd, 1 H, *J* = 4.8, 5.5 and 5.7, H-2′); 5.13 (t, 1 H, *J* = 5.4, 5′-OH); 5.25 (d, 1 H, *J* = 5.0, 3′-OH); 5.56 (d, 1 H, *J* = 5.7, 2′-OH); 6.11 (d, 1 H, *J* = 5.5, H-1′); 7.55−7.65 (m, 3 H, H−Ph); 8.84 (d, 2 H, *J* = 8.0, H−Ph); 8.93 (s, 1 H, H-8); 9.02 (s, 1 H, H-2). UV (λ_{max} (ϵ)) methanol: 290 (18300); water: pH 7, 289 (16800); pH 2, 298 (15700); pH 11, 289 (17100). Anal. ($C_{16}H_{16}N_4O_4$) C, H, N.

6-(4-Fluorophenyl)-9-(β-D-ribofuranosyl)purine (12b). Colorless crystals, yield 83%, mp 207−210 °C; $[\alpha]_D$ −51.9 (*c* 0.5 DMF). FAB MS *m/z* (rel. %): 347 (6) [M + H], 215 (7) [M + H − Rf], 185 (40), 93 (100). ^{1}H NMR (400 MHz, $CDCl_3$): 3.58−3.64 and 3.70−3.75 (2 × m, 2 H, CH_2-5′); 4.01 (m, 1 H, H-4′); 4.23 (m, 1 H, H-3′); 4.66 (ddd, 1 H, *J* = 4.9, 5.5 and 5.9, H-2′); 5.13 (t, 1 H, *J* = 5.5, 5′-OH); 5.24 (d, 1 H, *J* = 5.1, 3′-OH); 5.55 (d, 1 H, *J* = 5.9, 2′-OH); 6.10 (d, 1 H, *J* = 5.5, H-1′); 7.46 (t, 2 H, *J* = 8.9, H-*o*-Ar); 8.89−8.94 (m, 2 H, H-*m*-Ar); 8.94 (s, 1 H, H-8); 9.01 (s, 1 H, H-2). ^{13}C NMR (100 MHz, DMSO-d_6): 63.75 (CH_2-5′); 71.33 (CH-3′); 73.88 (CH-2′); 85.78 (CH-4′); 87.86 (CH-1′); 115.89 (d, *J* = 21.4, *m*-CH−FPh); 130.71 and 131.77 (*i*-C−FPh and C-5); 131.97 (d, *J* = 8.7, *o*-CH−FPh); 145.11 (C-8); 151.96 (C-2); 152.01 and 152.31 (C-4 and C-6); 164.06 (d, ^1J(C,F) = 250.0, CF). ^{19}F NMR (376.5 MHz, $CDCl_3$): −108.65 (m, FPh). UV (λ_{max} (ϵ)) methanol: 294 (25400). Anal. ($C_{16}H_{15}FN_4O_4$) C, H, N.

6-(2,4-Difluorophenyl)-9-(β-D-ribofuranosyl)purine (12c). Colorless crystals, yield 91%, mp 89−92 °C; $[\alpha]_D$ −49.0 (*c* 0.5 DMF). FAB MS *m/z* (rel. %): 365 (20) [M + H], 233 (24) [M + H − Rf], 93 (100). ^{1}H NMR (400 MHz, DMSO-d_6): 3.57−3.63 and 3.68−3.75 (2 × m, 2 H, CH_2-5′); 4.01 (m, 1 H, H-4′); 4.23 (m, 1 H, H-3′); 4.69 (ddd, 1 H, *J* = 5.3, 5.7 and 5.9, H-2′); 5.10 (t, 1 H, *J* = 5.5, 5′-OH); 5.25 (d, 1 H, *J* = 5.0, 3′-OH); 5.56 (d, 1 H, *J* = 5.9, 2′-OH); 6.10 (d, 1 H, *J* = 5.7, H-1′); 7.0−7.35 (m, 1 H, H-5-Ph); 7.48 (ddd, 1 H, *J* = 2.3, 9.7 and 10.7, H-3-Ph); 8.06 (dt, 1 H, *J* = 8.5 and 6.7, H-6-Ar); 8.90 (s, 1 H, H-8); 9.05 (s, 1 H, H-2). ^{13}C NMR (100 MHz, DMSO-d_6): 61.21 (CH_2-5′); 70.27 (CH-3′); 73.69 (CH-2′); 85.72 (CH-4′); 87.67 (CH-1′); 104.83 (t, *J* = 25.9, CH-3-Ph); 111.91 (d, *J* = 21.7, CH-5-Ph); 120.12 (C-1-Ph); 131.96 (C-5); 133.65 (d, *J* = 6.7, CH-6-Ph); 145.25 (C-8); 151.22 (C-6); 151.62 (C-4); 151.92 (C-2); 160.35 (dd, *J* = 12.8 and 256.0, CF-2-Ph); 163.51 (dd, *J* = 11.8 and 250.3, CF-4-Ph). ^{19}F NMR (376.5 MHz, DMSO-d_6): −106.30 (pent, *J* = 7.8) and −107.30 (q, *J* = 9.6, F_2Ph). UV (λ_{max} (ϵ)) methanol: 280 (26300). Anal. ($C_{16}H_{14}F_2N_4O_4$) C, H, N.

6-(3,4-Difluorophenyl)-9-(β-D-ribofuranosyl)purine (12d). Colorless crystals, yield 93%, mp 189−191 °C; $[\alpha]_D$ −48.0 (*c* 0.5 DMF). FAB MS *m/z* (rel. %): 365 (47) [M + H], 233 (100) [M + H − Rf]. ^{1}H NMR (400 MHz, DMSO-d_6): 3.58−3.64 and 3.70−3.76 (2 × m, 2 H, CH_2-5′); 4.02 (m, 1 H, H-4′); 4.23 (m, 1 H, H-3′); 4.66 (ddd, 1 H, *J* = 4.7, 5.4 and 5.9, H-2′); 5.13 (t, 1 H, *J* = 5.4, 5′-OH); 5.25 (d, 1 H, *J* = 5.0, 3′-OH); 5.56 (d, 1 H, *J* = 5.9, 2′-OH); 6.11 (d, 1 H, *J* = 5.4, H-1′); 7.68 (q, 1 H, *J* = 8.6, H-arom); 7.71−8.83 (m, 2 H, H-arom); 8.97 (s, 1 H, H-8); 9.02 (s, 1 H, H-2). ^{13}C NMR (100 MHz, DMSO-d_6): 61.11 (CH_2-5′); 70.15 (CH-3′); 73.79 (CH-2′); 85.64 (CH-4′); 87.76 (CH-1′); 117.87 (d, *J* = 6.0, CH-5-Ph); 118.03 (CH-2-Ph); 126.57 (CH-6-Ph); 130.67 (C-1-Ph); 132.71 (C-5); 145.32 (C-8); 149.48 (dd, *J* = 12.8 and 244.2, CF−Ph); 150.30 (C-6); 151.2248 (dd, *J* = 12.2 and 250.0, CF−Ph); 151.80 (C-2); 152.32 (C-4). ^{19}F NMR (376.5 MHz, DMSO-d_6): −133.97 and −137.13 (2 × m, F_2Ph). UV (λ_{max} (ϵ)) methanol: 298 (36700). Anal. ($C_{16}H_{14}F_2N_4O_4$) C, H, N.

9-(β-D-Ribofuranosyl)-6-(2-tolyl)purine (12e). Colorless crystals, yield 91%, mp 79−81 °C; $[\alpha]_D$ −51.6 (*c* 0.5 DMF). FAB MS *m/z* (rel. %): 343 (12) [M + H], 211 (100) [M + H − Rf]. ^{1}H NMR (400 MHz, DMSO-d_6): 2.37 (s, 3 H, CH_3); 3.58−3.64 and 3.70−3.76 (2 × m, 2 H, CH_2-5′); 4.02 (m, 1 H, H-4′); 4.23 (m, 1 H, H-3′); 4.71 (m, 1 H, H-2′); 5.14 (brs, 1 H, 5′-OH); 5.28 (brs, 1 H, 3′-OH); 5.58 (d, 1 H, *J* = 5.6, 2′-OH); 6.11 (d, 1 H, *J* = 5.8, H-1′); 7.12−7.68 (m, 4 H, H−Ph); 8.86 (s, 1 H, H-8); 9.04 (s, 1 H, H-2). UV (λ_{max} (ϵ)) methanol: 274 (23000). Anal. ($C_{17}H_{18}N_4O_4$) C, H, N.

9-(β-D-Ribofuranosyl)-6-(4-tolyl)purine (12f). Colorless crystals, yield 76%, mp 226−229 °C; $[\alpha]_D$ −59.0 (*c* 0.5 DMF). FAB MS *m/z* (rel. %): 343 (57) [M + H], 211 (72) [M + H − Rf], 201 (10), 185 (34), 93 (100). ^{1}H NMR (400 MHz, DMSO-d_6): 3.31 (s, 3 H, CH_3); 3.57−3.64 and 3.70−3.76 (2 × m, 2 H, CH_2-5′); 4.01 (m, 1 H, H-4′); 4.23 (m, 1 H, H-3′); 4.66 (ddd, 1 H, *J* = 5.0, 5.5 and 5.9, H-2′); 5.12 (t, 1 H, *J* = 5.6, 5′-OH); 5.22 (d, 1 H, *J* = 5.1, 3′-OH); 5.53 (d, 1 H, *J* = 5.9, 2′-OH); 6.10 (d, 1 H, *J* = 5.5, H-1′); 7.42 (d, 2 H, *J* = 8.2, H−Ph); 8.76 (d, 2 H, *J* = 8.2, H−Ph); 8.90 (s, 1 H, H-8); 8.98 (s, 1 H, H-2). UV (λ_{max} (ϵ)) methanol: 301 (22700). Anal. ($C_{17}H_{18}N_4O_4$) C, H, N.

6-(4-Methoxyphenyl)-9-(β-D-ribofuranosyl)purine (12g). Colorless crystals, yield 91%, mp 173−175 °C; $[\alpha]_D$ −61.5 (*c* 0.5 DMF). FAB MS *m/z* (rel. %): 359 (9) [M + H], 227 (8) [M + H − Rf], 201 (16), 185 (40), 93 (100). ^{1}H NMR (400 MHz, DMSO-d_6): 3.61 (dd, 1 H, *J* = 3.8 and 12.0) and 3.73 (dd, 1 H, *J* = 3.8 and 12.0, CH_2-5′); 3.87 (s, 3 H, OCH_3); 4.01 (m, 1 H, H-4′); 4.23 (dd, 1 H, *J* = 3.6 and 4.9, H-3′); 4.66 (dd, 1 H, *J* = 4.9 and 5.5, H-2′); OH signals were exchanged; 6.09 (d, 1 H, *J* = 5.5, H-1′); 7.16 (d, 2 H, *J* = 8.9, H−Ph); 8.85 (d, 2 H, *J* = 8.9, H−Ph); 8.88 (s, 1 H, H-8); 8.94 (s, 1 H, H-2). ^{13}C NMR (100 MHz, DMSO-d_6): 55.51 (CH_3); 61.40 (CH_2-5′); 70.43 (CH-3′); 73.89 (CH-2′); 85.81 (CH-4′); 87.83 (CH-1′); 114.28 (d, *m*-CH−OCH_3Ph); 127.85 (*i*-C−OCH_3Ph); 130.36 (C-5); 131.32 (*o*-CH−OCH_3Ph); 144.51 (C-8); 151.98 (C-2); 152.07 (C-4); 152.96 (C-6); 161.88 (C−OCH_3). UV (λ_{max} (ϵ)) methanol: 314 (21500). Anal. ($C_{17}H_{18}N_4O_5$) C, H, N.

6-(3-Methoxyphenyl)-9-(β-D-ribofuranosyl)purine (12h). Colorless crystals, yield 71%, mp 141−143 °C; $[\alpha]_D$ −49.2 (*c* 0.5 DMF). FAB MS *m/z* (rel. %): 359 (93) [M + H], 227 (100) [M + H − Rf], 201 (75). ^{1}H NMR (400 MHz, DMSO-d_6): 3.58−3.64 and 3.70−3.75 (2 × m, 2 H, CH_2-5′); 3.88 (s, 3 H, OCH_3); 4.01 (m, 1 H, H-4′); 4.23 (m, 1 H, H-3′); 4.66 (ddd, 1 H, *J* = 5.1, 5.5 and 5.9, H-2′); 5.11 (t, 1 H, *J* = 5.5, 5′-OH); 5.22 (d, 1 H, *J* = 5.0, 3′-OH); 5.54 (d, 1 H, *J* = 5.9, 2′-OH); 6.11 (d, 1 H, *J* = 5.5, H-1′); 7.17 (dd, 1 H, *J* = 2.1 and 7.7, H−Ph); 7.53 (t, 1 H, *J* = 8.0, H−Ph); 8.42 (brs, 1 H, H−Ph); 8.47 (d, 1 H, *J* = 7.9, H−Ph); 8.93 (s, 1 H, H-8); 9.01 (s, 1 H, H-2). UV (λ_{max} (ϵ)) methanol: 291 (29700). Anal. ($C_{17}H_{18}N_4O_5$) C, H, N.

6-(4-Ethoxyphenyl)-9-(β-D-ribofuranosyl)purine (12i). Colorless crystals, yield 89%, mp 173−176 °C; $[\alpha]_D$ −57.5 (*c* 0.5 DMF). FAB MS *m/z* (rel. %): 373 (50) [M + H], 279 (63), 241 (100) [M + H − Rf]. ^{1}H NMR (500 MHz, DMSO-d_6): 1.38 (t, 3 H, *J* = 6.9, CH_3CH_2); 3.61 (dd, 1 H, *J* = 3.8 and 12.0) and 3.73 (dd, 1 H, *J* = 3.8 and 12.0, CH_2-5′); 4.01 (m, 1 H, H-4′); 4.15 (q, 2 H, *J* = 6.9, CH_2CH_3); 4.22 (dd, 1 H, *J* = 3.6 and 4.8, H-3′); 4.66 (dd, 1 H, *J* = 4.8 and 5.6, H-2′); OH signals were exchanged; 6.09 (d, 1 H, *J* = 5.6, H-1′); 7.15 (d, 2 H, *J* = 8.9, H−Ph); 8.84 (d, 2 H, *J* = 8.9, H−Ph); 8.88 (s, 1 H, H-8); 8.94 (s, 1 H, H-2). UV (λ_{max} (ϵ)) methanol: 315 (24100). Anal. ($C_{18}H_{20}N_4O_5$) C, H, N.

6-(4-Chlorophenyl)-9-(β-D-ribofuranosyl)purine (12j). Colorless crystals, yield 82%, mp 206−208 °C; $[\alpha]_D$ −55.5 (*c* 0.5 DMF). FAB MS *m/z* (rel. %): 363 (32) [M + H], 279 (100), 231 (81) [M + H − Rf]. ^{1}H NMR (400 MHz, DMSO-d_6): 3.58 (dd, 1 H, *J* = 3.7 and 12.0) and 3.71 (dd, 1 H, *J* = 3.8 and 12.0, CH_2-5′); 3.99 (m, 1 H, H-4′); 4.20 (dd, 1 H, *J* = 3.6 and 4.8, H-3′); 4.64 (dd, 1 H, *J* = 4.8 and 5.4, H-2′); ca. 5.1, ca. 5.2 and ca. 5.55 (3 × brs, 3 × OH); 6.08 (d, 1 H, *J* = 5.4, H-1′); 7.67 (d, 2 H, *J* = 8.5, H−Ph); 8.84 (d, 2 H, *J* = 8.5, H−Ph); 8.94 (s, 1 H, H-8); 9.00 (s, 1 H, H-2). UV (λ_{max} (ϵ)) methanol: 295 (26000). Anal. ($C_{16}H_{15}ClN_4O_4$) C, H, N, Cl.

2-Amino-6-phenyl-9-(β-D-ribofuranosyl)purine (13a). Colorless crystals, yield 64%, mp 184−186 °C; $[\alpha]_D$ −35.3 (*c* 0.3 DMF). FAB MS *m/z* (rel. %): 344 (11) [M + H], 212 (37) [M + H − Rf], 115 (100). ^{1}H NMR (400 MHz, DMSO-d_6): 3.57 (dd, 1 H, *J* = 4.0 and 11.9) and 3.67 (dd, 1 H, *J* = 4.0 and 11.9, CH_2-5′); 3.93 (m, 1 H, H-4′); 4.17 (dd, 1 H, *J* = 3.7 and 5.0, H-3′); 4.51 (dd, 1 H, *J* = 5.0 and 5.7, H-2′); OH signals were exchanged; 5.90 (d, 1 H, *J* = 5.7, H-1′); 6.55 (s, 2 H, NH_2); 7.51−7.58 (m, 3 H, H−Ph); 8.41 (s, 1 H, H-8); 8.68−8.74 (m, 2 H, H−Ph). UV (λ_{max} (ϵ)) methanol: 335 (9600), 248 (16700); water: pH 7, 327 (9200), 247 (13800); pH 2, 342 (11200), 259 (10100), 227 (23400); pH 11, 327 (9100), 247 (13800). Anal. ($C_{16}H_{17}N_5O_4$) C, H, N.

2-Amino-6-(4-fluorophenyl)-9-(β-D-ribofuranosyl)purine (13b). Yellowish crystals, yield 81%, mp 199−202 °C; $[\alpha]_D$ −29.1 (*c* 0.4 DMF). FAB MS *m/z* (rel. %): 362 (20) [M + H], 230 (22) [M + H − Rf], 93 (100). ^{1}H NMR (400 MHz, DMSO-d_6): 3.55−3.70 (m, 2 H, CH_2-5′); 3.93 (m, 1 H, H-4′); 4.16 (dd, 1 H, *J* = 3.6 and 4.7, H-3′); 4.55 (dd, 1 H, *J* = 4.7 and 5.9, H-2′); 5.13 (t, 1 H, *J* = 5.5, 5′-OH); 5.08, 5.14 and 5.44 (3 × brs, 3 × 1 H, 3 × OH); 5.90 (d, 1 H, *J* = 5.9, H-1′); 6.58 (s, 2 H, NH_2); 7.39 (t, 2 H, *J* = 8.9, H-*o*-Ar); 8.41 (s, 1 H, H-8); 8.79 (dd, 2 H, *J* = 8.9 and 5.8, H-*m*-Ar). ^{19}F NMR (376.5 MHz, DMSO-d_6): −109.62 (m, FPh). Anal. ($C_{16}H_{16}FN_5O_4$) C, H, N.

2-Amino-6-(2,4-difluorophenyl)-9-(β-D-ribofuranosyl)purine (13c). Yellowish crystals, yield 86%, mp 119−121 °C; $[\alpha]_D$ −24.6 (*c* 0.5 DMF). FAB MS *m/z* (rel. %): 380 (43) [M + H], 248 (60) [M + H − Rf], 93 (100). ^{1}H NMR (400 MHz, DMSO-d_6): 3.53−3.60 and 3.63−3.69 (2 × m, 2 H, CH_2-5′); 3.93 (m, 1 H, H-4′); 4.15 (m, 1 H, H-3′); 4.55 (ddd, 1 H, *J* = 4.9, 6.0 and 6.0, H-2′); 5.05 (t, 1 H, *J* = 5.4, 5′-OH); 5.16 (d, 1 H, *J* = 4.7, 3′-OH); 5.46 (d, 1 H, *J* = 6.0, 2′-OH); 5.89 (d, 1 H, *J* = 6.0, H-1′); 6.66 (s, 2 H, NH_2); 7.26 (dt, 1 H, *J* = 2.5 and 8.4, H-arom); 7.40 (dt, 1 H, *J* = 2.4 and 10.0, H-arom); 7.87 (q, 1 H, *J* = 7.1, H-arom); 8.34 (s, 1 H, H-8). ^{13}C NMR (100 MHz, DMSO-d_6): 61.38 (CH_2-5′); 70.41 (CH-3′); 73.39 (CH-2′); 85.37 (CH-4′); 86.39 (CH-1′); 104.57 (t, *J* = 25.9, CH-3-Ph); 111.91 (d, *J* = 21.1, CH-5-Ph); 120.71 (C-1-Ph); 125.26 (C-5); 133.12 (d, *J* = 9.5, CH-6-Ph); 140.82 (C-8); 152.29 (C-6); 153.82 (C-4); 160.27 (C-2); 160.11 (dd, *J* = 13.0 and 254.9, CF-2-Ph); 163.16 (dd, *J* = 12.4 and 249.6, CF-4-Ph). ^{19}F NMR (376.5 MHz, DMSO-d_6): −107.33 − −107.45 (m, F_2Ph). Anal. ($C_{16}H_{15}F_2N_5O_4$) C, H, N.

2-Amino-6-(3,4-difluorophenyl)-9-(β-D-ribofuranosyl)purine (13d). Hygroscopic yellowish solid, yield 83%, mp 181−185 °C; $[\alpha]_D$ −19.9 (*c* 0.4 DMF). FAB MS *m/z* (rel. %): 380 (20) [M + H], 248 (100) [M + H − Rf], 93 (77). ^{1}H NMR (400 MHz, DMSO-d_6): 3.55−3.70 (m, 2 H, CH_2-5′); 3.94 (m, 1 H, H-4′); 4.16 (dd, 1 H, *J* = 3.3 and 4.9, H-3′); 4.54 (dd, 1 H, *J* = 4.9 and 5.8, H-2′); signals of OH groups were exchanged; 5.90 (d, 1 H, *J* = 5.8, H-1′); 6.65 (brs, 2 H, NH_2); 7.64 (q, 1 H, *J* = 8.8, H−Ar); 8.45 (s, 1 H, H-8); 8.61−8.70 (m, 2 H, H-*m*-Ar). ^{19}F NMR (376.5 MHz, DMSO-d_6): −134.96 and −137.72 (2 × m, F_2Ph). Anal. ($C_{16}H_{15}F_2N_5O_4 \cdot H_2O$) C, H. N.

Inhibition of Cell Growth. Mouse leukemia L1210 cells (ATCC CCL 219) were cultivated in RPMI 1640 medium containing 15% bovine serum[38] using 24-well tissue culture plates. The cells were seeded at 5×10^4 mL^{-1} and after a 24-h incubation period (CO_2 atmosphere, 37 °C) tested compounds were added at five different concentrations. The endpoint of the cell growth was 72 h following the addition. An appropriate aliquot from every dish was then counted (cell counter Serono 150+). The inhibitory potency of the compounds tested was expressed as IC_{50} values.

CCRF-CEM T-lymphoblastoid cells (ATCC CCL 119) were cultivated in RPMI 1640 medium supplemented with L-glutamine (0.3 g/L) containing 10% bovine serum using 24-well tissue culture plates. The cells were seeded at 10^5 mL^{-1} and after a 24-h incubation period (CO_2 atmosphere, 37 °C) tested compounds were added at five different concentrations. The endpoint of the cell growth was 72 h following the drug addition. An appropriate aliquot from every dish was then counted (cell counter Serono 150+). The inhibitory potency of the compounds tested was expressed as IC_{50} values.

Murine L929 cells (ATCC CCL 1) were placed in 24-well tissue dishes and grown in Waymouth's MB 752/1 medium containing 5% fetal calf serum (10^5 cells/dish). After 24 h cell monolayers were overlaid with fresh medium supplemented with tested compounds at five different concentrations. After the additional 48-h incubation at 37 °C (CO_2 atmosphere) cultures were stained with methylene blue (Sigma)[39] and the absorbance at 600 nm was then measured in 1% sarkosyl (Sigma) extracts. The number of cells was calculated from the calibration curve and results were expressed as IC_{50} values.

Human cervix carcinoma HeLa S3 cells (ATCC CCL 2.2) were seeded in 24-well dishes (RPMI 1640 HEPES modification containing 5% fetal bovine serum) (7×10^4 cells/mL) and incubated 24 h at 37 °C (CO_2 atmosphere). Then the culture medium was removed and fresh medium with test compound was added at five different concentrations; 48 h following drug addition cultivation was stopped and the cell growth was evaluated after staining with methylene blue (Sigma)[39] (see above).

Cell Viability. In parallel, the number of viable cells (viability in cell population) was quantified using MTT standard spectrophotometric assay.[40]

Acknowledgment. This work was supported by the Grant Agency of the Czech Republic (Grants 203/98/P027 and 203/96/K001). The excellent technical assistance of Ms. Kamila Havlíčková is gratefully acknowledged. The authors also thank the staff of the Mass Spectrometry and Analytical Departments of this Institute.

References

(1) (a) Robins, R. K.; Revankar, G. R. Purine analogues and related nucleosides and nucleotides as antitumor agents. *Med. Res. Rev.* **1985**, *5*, 273−296. (b) Plunkett, W.; Saunders, P. P. Metabolism and action of purine nucleoside analogues. *Pharmacol. Ther.* **1991**, *49*, 239−268. (c) Cheson, B. D. Perspectives on purine analogues. *Hematol. Cell Ther.* **1996**, *38* (Suppl. 2), S109−S116. (d) Bergmann, L. Present status of purine analogues in the therapy of chronic lymphocytic leukemias. *Leukemia* **1997**, *11* (Suppl. 2), S29−S34.

(2) (a) Veselý, J.; Havlíček, L.; Strnad, M.; Blow, J. J.; Donella-Deana, A.; Pinna, L.; Letham, D. S.; Kato, J.; Detivaud, L.; Leclerc, S. Inhibition of cyclin-dependent kinases by purine analogues. *Eur. J. Biochem.* **1994**, *224*, 771−786. (b) Havlíček, L.; Hanus, J.; Veselý, J.; Leclerc, S.; Meijer, L.; Shaw, G.; Strnad, M. Cytokinin-derived cyclin-dependent kinase inhibitors: synthesis and cdc2 inhibitory activity of olomoucine and related compounds. *J. Med. Chem.* **1997**, *40*, 408−412. (c) Legraverend, M.; Ludwig, O.; Bisagni, E.; Leclerc, S.; Meijer, L. Synthesis of C2 alkynylated purines, a new family of potent inhibitors of cyclin-dependent kinases. *Bioorg. Med. Chem. Lett.* **1998**, *8*, 793−798. (d) Legraverend, M.; Ludwig, O.; Bisagni, E.; Leclerc, S.; Meijer, L.; Giocanti, N.; Sadri, R.; Favaudon, V. Synthesis and in vitro evaluation of novel 2,6,9-trisubstituted purines acting as cyclin-dependent kinase inhibitors. *Bioorg. Med. Chem.* **1999**, *7*, 1281−1293. (e) N.Oh, C. H.; Lee, S. C.; Lee, K. S.; Woo, E. R.; Hong, C. Y.; Yang, B. S.; Baek, D. J.; Cho, J. H. Synthesis and biological activities of C-2, N-9 substituted 6-benzylaminopurine derivatives as cyclin-dependent kinase inhibitors. *Arch. Pharm. (Weinheim)* **1999**, *332*, 187−190.

(3) Revankar, G. R.; Robins, R. K. Heterocyclic Analogues of Purine Nucleosides and Nucleotides. In *Chemistry of Nucleosides and Nucleotides*; Townsend, L. B., Ed.; Plenum Press: New York/London, 1988; Vol. 2, pp 200−246.

(4) (a) Cristalli, G.; Franchetti, P.; Grifantini, M.; Vittori, S.; Bordoni, T.; Geroni, C. Improved synthesis and antitumor activity of 1-deazaadenosine. *J. Med. Chem.* **1987**, *30*, 1686−1688. (b) Cristalli, G.; Vittori, S.; Eleuteri, A.; Grifantini, M.; Volpini, R.; Lupidi, G.; Capolongo, L.; Pesenti, E. Purine and 1-deazapurine ribonucleosides and deoxyribonucleosides: synthesis and biological activity. *J. Med. Chem.* **1991**, *34*, 2226−2230.

(5) Franchetti, P.; Cappellacci, L.; Grifantini, M.; Lupidi, G.; Nocentini, G.; Barzi, A. 8-Aza Analogues of Deaza Purine Nucleosides − Synthesis and Biological Evaluation of 8-Aza-1-deazaadenosine and 2′-Deoxy-8-aza-1-deazaadenosine. *Nucleosides Nucleotides* **1992**, *11*, 1059−1076.

(6) (a) Leopold, W. R.; Fry, D. W.; Boritzki, T. J.; Besserer, J. A.; Pattison, I. C.; Jackson, R. C. Deazaguanine mesylate: a new antipurine antimetabolite. *Invest. New Drugs* **1985**, *3*, 223−231.

(7) (a) Page, T.; Jacobsen, S. J.; Smejkal, R. M.; Scheele, J.; Nyhan, W. L.; Mangum, J. H.; Robins, R. K. Studies on the mechanism of cytotoxicity of 3-deazaguanosine in human cancer cells. *Cancer Chemother. Pharmacol.* **1985**, *15*, 59−62. (b) Jacobsen, S. J.; Page, T.; Diala, E. S.; Nyhan, W. L.; Robins, R. K.; Mangum, J. H. Synergistic activity of purine metabolism inhibitors in cultured human tumor cells. *Cancer Lett.* **1987**, *35*, 97−104.

(8) Pudlo, J. S.; Nassiri, M. R.; Kern, E. R.; Wotring, L. L.; Drach, J. C.; Townsend, L. B. Synthesis, antiproliferative, and antiviral activity of certain 4-substituted and 4,5-disubstituted 7-[(1,3-dihydroxy-2-propoxy)methyl]pyrrolo[2,3-d]pyrimidines. *J. Med. Chem.* **1990**, *33*, 1984−1992.

(9) Elion, G. B.; Hitchings, G. H. Metabolic basis for the actions of analogues of purines and pyrimidines. *Adv. Chemother.* **1965**, *2*, 91−177.

(10) For review: Benezra, S. A.; Foss P. R. B. In *Analytical Profiles of Drug Substances*;Florey, K., Ed.; Academic Press: New York, 1978; pp 343−357.

(11) (a) Avery, T. L.; Finch, R. A.; Vasquez, K. M.; Radparvar, S.; Hanna, N. B.; Revankar, G. R.; Robins, R. K. Chemotherapeutic characterization in mice of 2-amino-9-beta-D-ribofuranosylpurine-6-sulfinamide (sulfinosine), a novel purine nucleoside with unique antitumor properties. *Cancer Res.* **1990**, *50*, 2625−2630. (b) Revankar, G. R.; Hanna, N. B.; Imamura, N.; Lewis, A. F.; Larson, S. B.; Finch, R. A.; Avery, T. L.; Robins, R. K. Synthesis and in vivo antitumor activity of 2-amino-9H-purine-6-sulfenamide, -sulfinamide, and -sulfonamide and related purine ribonucleosides. *J. Med. Chem.* **1990**, *33*, 121−128. (c) Hanna, N. B.; Bhattacharya, B. K.; Robins, R. K.; Avery, T. L.; Revankar, G. R. Sulfinosine Congeners − Synthesis and Antitumor Activity in Mice of Certain N9−Alkylpurines and Purine Ribonucleosides. *J. Med. Chem.* **1994**, *37*, 177−183.

(12) Williams, B. A.; Blay, J.; Hoskin, D. W. 2-chloroadenosine stimulates granule exocytosis from mouse natural killer cells: Evidence for signal transduction through a novel extracellular receptor. *Exp. Cell Res.* **1997**, *233*, 187−197.

(13) (a) Wataya, Y.; Hirota, Y.; Hiramoto-Yoshioka, A.; Tanaka, S.; Otani, T.; Minowada, J.; Matsuda, A.; Ueda, T. The mechanism of 2-chlorodeoxyadenosine-induced cell death. *Adv. Exp. Med. Biol.* **1989**, *253B*, 227−234. (b) Carson, D. A.; Wasson, D. B.; Taetle, R.; Yu, A. Specific toxicity of 2-chlorodeoxyadenosine toward resting and proliferating human lymphocytes. *Blood* **1983**, *62*, 737−743. (c) Carson, D. A.; Wasson, D. B.; Beutler, E. Antileukemic and immunosuppressive activity of 2-chloro-2′-deoxyadenosine. *Proc. Natl. Acad. Sci. U.S.A.* **1984**, *81*, 2232−2236.

(14) (a) Reiter, Z.; Tomson, S.; Ozes, O. N.; Taylor, M. W. Combination treatment of 2-chlorodeoxyadenosine and type I interferon on hairy cell leukemia-like cells: cytotoxic effect and MHC-unrestricted killer cell regulation. *Blood* **1993**, *81*, 1699−1708. (b) Saven, A.; Carrera, C. J.; Carson, D. A.; Beutler, E.; Piro, L. D. 2-Chlorodeoxyadenosine: an active agent in the treatment of cutaneous T-cell lymphoma. *Blood* **1992**, *80*, 587−592. (c) Arner, E. S. J. On the phosphorylation of 2-chlorodeoxyadenosine (CdA) and its correlation with clinical response in leukemia treatment. *Leuk. Lymphoma* **1996**, *21*, 225−231.

(15) (a) Keating, M. J.; Kantarian, H.; Talpaz, M. Fludarabine: A new agent with major activity against chronic lymphocytic leukemia. *Blood* **1989**, *74*, 19−25. (b) Pawelski, S. Fludarabine in the treatment of chronic lymphocytic leukemia and other lymphoproliferative disorders. *Acta Haematol. Pol.* **1995**, *26*, 263−268. (c) Fludarabine phosphate: an effective therapy for lymphoid malignancies. *Semin. Oncol.* **1990**, *17*, 1−78.

(16) Matsuda, A.; Shinozaki, M.; Yamaguchi, T.; Homma, H.; Nomoto, R.; Miyasaka, T.; Watanabe, Y.; Abiru, T. Nucleosides and Nucleotides. 103. 2-Alkynyladenosines − A Novel Class of Selective Adenosine-A2 Receptor Agonists with Potent Antihypertensive Effects. *J. Med. Chem.* **1992**, *35*, 241−252.

(17) Lambe, C. U.; Averett, D. R.; Paff, M. T.; Reardon, J. E.; Wilson, J. G.; Krenitsky, T. A. 2-Amino-6-methoxypurine arabinoside: an agent for T-cell malignancies. *Cancer Res.* **1995**, *55*, 3352−3356.

(18) (a) Gibboney, D. S.; French, B. T.; Patrick, D. E.; Trewyn, R. W. 6-ethylmercaptopurine-mediated growth inhibition of HL-60 cells in vitro irrespective of purine salvage. *Cancer Chemother. Pharmacol.* **1989**, *25*, 189−194. (b) Akiyama, S. I.; Kuwano, M.; Komiyama, S.; Saneyoshi, M. Antitumor effect of a combination of 6-methylthioinosine and amphotericin B on mouse leukemia L1210. *Cancer Lett.* **1980**, *9*, 305−311.

(19) (a) McGarrity, G. J.; Carson, D. A. Adenosine phosphorylase-mediated nucleoside toxicity. Application towards the detection of mycoplasmal infection in mammalian cell cultures. *Exp. Cell Res.* **1982**, *139*, 199−205. (b) Whitaker, A. M.; Windsor, G. D.; Burnett, C. M.; Taylor, C. H. A rapid and sensitive method for the detection of mycoplasmas in infected cell cultures using 6-methylpurine deoxyriboside. *Dev. Biol. Stand.* **1987**, *66*, 503−509. (c) Ishiguro, K.; Taira, S.; Sasaki, T.; Nariuchi, H. Depletion of mycoplasma from infected cell lines by limiting dilution in 6-methylpurine deoxyriboside. *J. Immunol. Methods* **1988**, *108*, 39−43.

(20) (a) Parker, W. B.; King, S. A.; Allan, P. W.; Bennett, L. L., Jr.; Secrist, J. A.; Montgomery, J. A.; Gilbert, K. S.; Waud, W. R.; Wells, A. H.; Gillespie, G. Y.; Sorscher, E. J. In vivo gene therapy of cancer with E. coli purine nucleoside phosphorylase. *Hum. Gene Ther.* **1997**, *8*, 1637−1644. (b) Parker, W. B.; Allan, P. W.; Shaddix, S. C.; Rose, L. M.; Speegle, H. F.; Gillespie, G. Y.; Bennett, L. L., Jr. Metabolism and metabolic actions of 6-methylpurine and 2-fluoroadenine in human cells. *Biochem. Pharmacol.* **1998**, *55*, 1673−1681.

(21) Brathe, A.; Gundersen, L. L.; Rise, E.; Eriksen, A. B.; Vollsnes, A. V.; Wang, L. N. Synthesis of 6-alkenyl- and 6-alkynylpurines with cytokinin activity. *Tetrahedron* **1999**, *55*, 211−228.
(22) (a) Kobayashi, Y.; Yamamoto, K.; Asai, T.; Nakano, M.; Kumadaki, I. Studies on Organic Fluorine Compounds. Part 35. Trifluoromethylation of Pyrimidine- and Purine-nucleosides with Trifluoromethyl-Copper Complex. *J. Chem. Soc., Perkin Trans. 1* **1980**, 2755−2761. (b) Hocková, D.; Hocek, M.; Dvořáková, H.; Votruba, I. Synthesis and Cytostatic Activity of Nucleosides and Acyclic Nucleoside Analogues Derived form 6-(Trifluoromethyl)-purines. *Tetrahedron* **1999**, *55*, 11109−11118.
(23) Cocuzza, A. J.; Chidester, D. R.; Culp, S.; Fitzgerald, L.; Gilligan, P. Use of Suzuki Reaction for the Synthesis of Aryl-substituted Heterocycles as Corticotropin-Releasing Hormone (CHR) Antagonists. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 1063−1066.
(24) (a) Moran, S.; Ren, R. X. F.; Rumney, S., IV; Kool, E. T. Difluorotoluene, A Nonpolar Isostere for Thymine, Codes Specifically and Efficiently for Adenine in DNA Replication. *J. Am. Chem. Soc.* **1997**, *119*, 2056−2057. (b) Liu, D.; Moran, S.; Kool, E. T. Bi-stranded, multisite replication of a base pair between difluorotoluene and adenine: confirmation by ′inverse′ sequencing. *Chem. Biol.* **1997**, *4*, 919−926 (c) Guckian, K. M.; Krugh, T. R.; Kool, E. T. Solution structure of a DNA duplex containing a replicable difluorotoluene-adenine pair. *Nat. Struct. Biol.* **1998**, *5*, 954−959.
(25) (a) Bergstrom, D. E.; Reddy, P. A. Synthesis of 6-Alkyl and 6-Aryl Substituted 9-β-D-ribofuranosyl Purines via the Nickel Catalyzed Coupling of Grignard Reagents to 2′,3′,5′-Tris-O-(*tert*-butyldimethylsilyl)-9-β-D-ribofuranosyl-6-chloropurine. *Tetrahedron Lett.* **1982**, *23*, 4191−4194. (b) Estep, K. G.; Josef, K. A.; Bacon, E. R.; Carabates, P. M.; Rumney, S., IV; Pilling, G. M.; Krafte, D. S.; Volberg, W. A.; Dillon, K.; Dugrenier, N.; Briggs, G. M.; Canniff, P. C.; Gorczyca, W. P.; Stankus, G. P.; Ezrin, A. M. Synthesis and Structure−Activity Relationship of 6-Heterocyclic-Substituted Purines as Inactivation Modifiers of Cardiac Sodium Channels. *J. Med. Chem.* **1995**, *38*, 2582−2595.
(26) (a) Gundersen, L. L. 6-Chloropurines and Organostannanes in Paladium Catalyzed Cross Coupling Reactions. *Tetrahedron Lett.* **1994**, *35*, 3155−3158. (b) Gundersen, L. L.; Bakkestuen, A. K.; Aasen, A. J.; Øveras, H.; Rise, F. 6-Halopurines in Palladium-Catalyzed Coupling with Organotin and Organozinc Reagents. *Tetrahedron* **1994**, *50*, 9743−9756. (c) Hocek, M.; Masojídková, M.; Holý, A. Synthesis of Acyclic Nucleotide Analogues Derived from *N*-substituted 6-(1-Aminomethyl)purines via 6-Acetylpurine Derivatives. *Tetrahedron* **1997**, *53*, 2291−2302. (d) Hocek, M.; Masojídková, M.; Holý, A. Synthesis of Acyclic Nucleotide Analogues Derived from 6-Hetarylpurines via Cross-Coupling Reactions of 9-[2-(Diethoxyphosphonylmethoxy)ethyl]-6-iodopurine with Hetaryl Organometallic Reagents. *Collect. Czech. Chem. Commun.* **1997**, *62*, 136−146. (e) Van Aerschot, A. A.; Mamos, P.; Weyns, N. J.; Ikeda, S.; De Clercq, E.; Herdewijn, P. Antiviral Activity of *C*-Alkylated Purine Nucleosides Obtained by Cross-Coupling with Tetraalkyltin Reagents. *J. Med. Chem.* **1995**, *36*, 2938−2942.
(27) Hirota, K.; Kitade, Y.; Kanbe, Y.; Maki, Y. Convenient Method for the Synthesis of *C*-Alkylated Purine Nucleosides: Palladium-Catalyzed Cross-Coupling Reaction of Halopurine Nucleosides with Trialkylaluminums. *J. Org. Chem.* **1992**, *57*, 5268−5270.
(28) (a) McKenzie, T. C.; Glass, D. The Reaction of 6-Halopurines with Phenyl Metal Complexes. *J. Heterocycl. Chem.* **1987**, *24*, 1551−1553. (b) Dvořáková, H.; Dvořák, D.; Holý, A. Coupling of 6-Chloropurines with Organocuprates Derived from Grignard Reagents: A Convenient Route to sec- and tert- 6-Alkylpurines. *Tetrahedron Lett.* **1996**, *37*, 1285−1288. (c) Dvořáková, H.; Dvořák, D.; Holý, A. Synthesis of Acyclic Nucleotide Analogues Derived from 6-(sec- or tert-Alkyl)purines via Coupling of 6-Chloropurine Derivatives with Organocuprates. *Collect. Czech. Chem. Commun.* **1998**, *63*, 2065−2074. (d) Hocek, M.; Holý, A. Perfluoroalkylation of 6-Iodopurines by Trimethyl(perfluoroalkyl)silanes. Synthesis of 6-(Perfluoroalkyl)purine Bases, Nucleosides and Acyclic Nucleotide Analogues. *Collect. Czech. Chem. Commun.* **1999**, *64*, 229−241.
(29) (a) Stevenson, T. M.; Prasad, A. S. B.; Citineni, J. B.; Knochel, P. Preparation of Zinc Organometallics Derived from Nucleosides and Nucleic Acid Bases and Pd(0) Catalyzed Coupling with Aryl Iodides. *Tetrahedron Lett.* **1996**, *37*, 8375−8378. (b) Prasad, A. S. B.; Stevenson, T. M.; Citineni, J. B.; Nyzam, V.; Knochel, P. Preparation and Reactions of New Zincated Nitrogen-Containing Heterocycles. *Tetrahedron* **1997**, *53*, 7237−7254.
(30) (a) Nair, V.; Richardson, S. G.; Coffman, R. E. Arylation and Heteroarylation of Photochemically Generated Purinyl Radicals. *J. Org. Chem.* **1982**, *47*, 4520−4524. (b) McKenzie, T. C.; Epstein, J. W. Coupling of Diazopurines, a Curious Steric Effect in a Free Radical Reaction. *J. Org. Chem.* **1982**, *47*, 4881−4884. (c) Nair, V.; Young, D. A. Synthetic Transformations of Transient Purinyl Radicals: Formation of Mono- and Diarylated and Heteroarylated Nucleosides. *J. Org. Chem.* **1984**, *49*, 4340−4344.
(31) Havelková, M.; Hocek, M.; Česnek, M.; Dvořák, D. The Suzuki-Miyaura Cross-Coupling Reactions of 6-Halopurines with Boronic Acids Leading to 6-Aryl- and 6-Alkenylpurines. *Synlett* **1999**, 1145−1147.
(32) Robins, R. K.; Godefroi, E. F.; Taylor, E. C.; Lewis, L. R.; Jackson, A. Purine Nucleosides. I. The Synthesis of Certain 6-Substituted-9-(tetrahydro-2-pyranyl)-purines as Model of Purine Deoxynucleotides. *J. Am. Chem. Soc.* **1961**, *83*, 2574−2579.
(33) Hocek, M.; Holý, A. A Facile Synthesis of 6-Cyanopurine Bases. *Collect. Czech. Chem. Commun.* **1995**, *60*, 1386−1389.
(34) Buck, I. M.; Reese, C. B. An Unambiguous Synthesis of Adenylsuccinic Acid and its Constituent Nucleoside. *J. Chem. Soc., Perkin Trans. 1* **1990**, 2937−2942.
(35) Robins, M. J.; Uznanski, B. Nucleic Acids Related Compounds. 33. Conversions of Adenosine and Guanosine to 2,6-Dichloro, 2-Amino-6-chloro, and Derived Purine Nucleosides. *Can. J. Chem.* **1981**, *59*, 2601−2607.
(36) Sugimura, H.; Takei, H. Synthesis of 6-Alkylpurine Derivatives by Nickel-Complex-Catalyzed Coupling Reaction of 6-(Methylthio)purine derivatives with Grignard Reagents. *Bull. Chem. Soc. Jpn.* **1985**, *58*, 664−666.
(37) Kos, N. J.; van der Plas, H. C. Occurrence of the S_NANRORC Mechanism in the Amination of 2-Substituted Purines with Potassium Amide in Liquid Ammonia. *J. Org. Chem.* **1980**, *45*, 2942−2945.
(38) Veselý, J. Synergistic effect of cis-dichlorodiammineplatinum and 5-aza-2′-deoxycytidine on mouse leukemic cells in vivo and in vitro. *Int. J. Cancer* **1982**, *29*, 81−85.
(39) Lillie, R. D. *H. J. Conn's Biological Stains*, 9th ed.; Williams & Wilkins: Baltimore, 1977.
(40) Carmichael, J.; DeGraff, W. G.; Gazdar, A. F.; Minna, J. D.; Mitchell J. B. Evaluation of a tetrazolium-based semiautomated colorimetric assay: assessment of chemosensitivity testing. *Cancer Res.* **1987**, *47*, 936−942.

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