Synthesis and Antiviral/Antitumor Activities of Certain 3-Deazaguanine Nucleosides and Nucleotides

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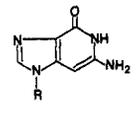
A new procedure for the preparation of the antiviral and antitumor agent 3-deazaguanine (1) and its metabolite 3-deazaguanosine (2) has been developed by reacting methyl 5(4)-(cyanomethyl)imidazole-4(5)-carboxylate (4) and 5-(cyanomethyl)-1-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)imidazole-4-carboxylate (6), respectively, with hydrazine. The 3-deazaguanosine 3′,5′-cyclic phosphate (13) was prepared from 5-(cyanomethyl)-1-β-D-ribofuranosylimidazole-4-carboxamide 5′-phosphate. Glycosylation of the trimethylsilyl 4 with 1-*O*-methyl-2-deoxy-3,5-di-*O*-*p*-toluoyl-D-ribofuranose in the presence of trimethylsilyl trifluoromethanesulfonate gave the corresponding N-1 and N-3 glycosyl derivatives with α-configuration (18 and 20) as the major products, along with minor amounts of the β-anomers (19 and 21). However, glycosylation of the sodium salt of 4 with 1-chloro-2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentofuranose (17) gave exclusively the β-anomers (19 and 21) in good yield. Base-catalyzed ring closure of these imidazole nucleosides gave 2′-deoxy-3-deazaguanosine (29), the α-anomer 28, and the corresponding N-3 positional isomers 27 and 26. The site of glycosylation and the anomeric configuration of these nucleosides have been assigned on the basis of ^{1}H NMR and UV spectral characteristics and by single-crystal X-ray analysis for 27–29. In a preliminary screening, several of these compounds have demonstrated significant broad-spectrum antiviral activity against certain DNA and RNA viruses in vitro, as well as moderate activity against L1210 and P388 leukemia in cell culture.

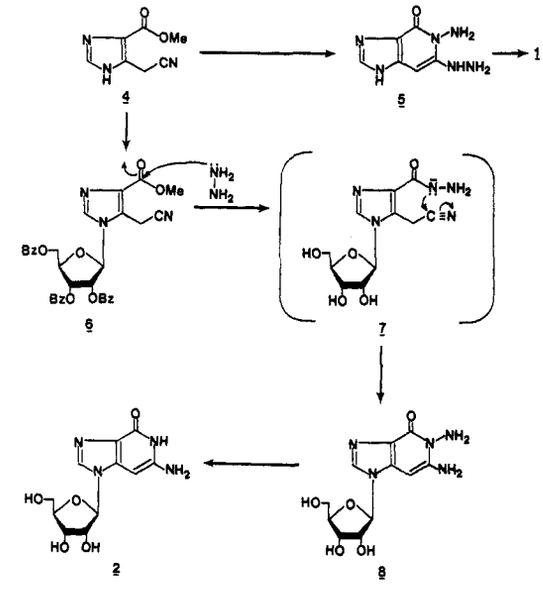
6-Amino-1,5-dihydroimidazo[4,5-*c*]pyridin-4-one (3-deazaguanine, 1) is a structural analogue of guanine in which the pyrimidine ring nitrogen in the 3-position is replaced by CH and was first reported by us[1] in 1976. Since that report,[1] the broad-spectrum antiviral activity against a variety of DNA and RNA viruses,[2] as well as the potent antitumor activity against L1210 leukemia and mammary adenocarcinomas in mice[3-5] of 1 and its metabolites 3-deazaguanosine (2, 6-amino-1,5-dihydro-1-β-D-ribofuranosylimidazo[4,5-*c*]pyridin-4-one) and 3-deazaguanylic acid (3, 6-amino-1,5-dihydro-1-β-D-ribofuranosylimidazo-[4,5-*c*]pyridin-4-one 5′-phosphate), have been confirmed independently in a number of laboratories. 3-Deazaguanine (1) exhibited potent cytotoxic activity in vitro against a variety of tumor cells, which include L1210,[3,5,6] HeLa,[3] human KB cells,[7] Ehrlich ascites tumor cells,[7] Chinese hamster ovary cells,[8] primary Chinese hamster embryo cells,[4] and mammary carcinoma EMT-6 cells.[9]



1, R = H
2, R = β-D-ribofuranose
3, R = β-D-ribofuranosyl 5′-phosphate

3-Deazaguanine has shown inhibitory action against a broad spectrum of animal breast tumor models, including rat mammary adenocarcinomas, mouse mammary adenocarcinomas,[4,5] slow- and fast-growing mammary tumors in mice, and the human breast xenograft subrenal capsule implant system. These observations are very interesting, since mammary adenocarcinoma (e.g., R3230AC) is a nonhormone-dependent estrogen-sensitive slow-growing tumor model for postoperative breast carcinomas and is not particularly susceptible to conventional cancer chemotherapeutic agents. Mammary adenocarcinoma 13762 is a fast-growing nonhormone-dependent tumor which is not responsive to estrogen treatment.[10] These results suggest a possible use of 3-deazaguanine in chemotherapy

Scheme I



of human breast cancer. 3-Deazaguanine also inhibited the growth of *Escherichia coli* B3 in vitro but did not

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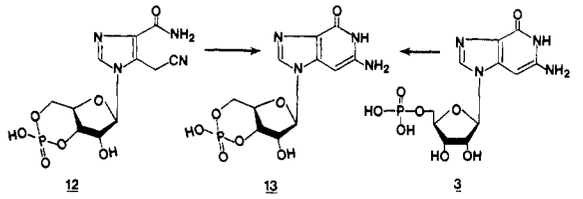
Scheme II



12 13 3

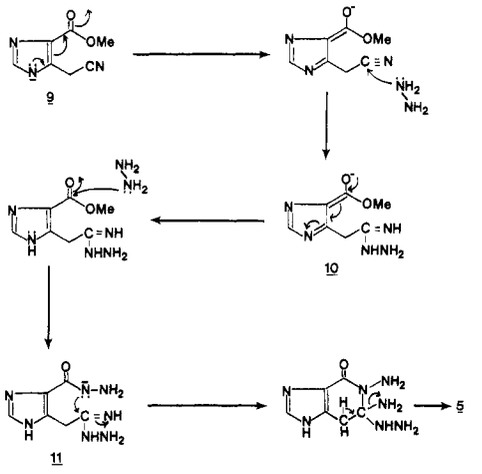
exhibit any antibacterial activity in vivo.[11,12] The 7-ribosyl derivative of **1** shows antibacterial activity against several Gram-negative strains in vivo without any appreciable toxicity to the host.[11] The antibacterial action of 7-ribosyl-3-deazaguanine has been ascribed to its cleavage to **1** in *E. coli B* infected cells.[13] Recently, 3-deazaguanosine (**2**) was shown to be a potent antileishmanial agent,[14] which is at least 20 times more active than **1** or allopurinol ribonucleoside against *Leishmania tropica* in vitro but less active than formycin B.

These unusual biological activities of **1** and **2** necessitated larger quantities of these compounds for further studies. We now report a convenient synthesis of **1** and **2** by a new ring-closure procedure along with the synthesis of 3-deazaguanosine 3′,5′-cyclic phosphate (**13**), 2′-deoxy-3-deazaguanosine (6-amino-1,2-dihydro-1-(2-deoxy-β-D-*erythro*-pentofuranosyl)imidazo[4,5-*c*]pyridin-4-one, **29**), and its positional isomer 7-(2-deoxy-β-D-*erythro*-pentofuranosyl)-3-deazaguanine (**27**).

Chemistry. The starting material, methyl 5(4)-(cyanomethyl)imidazole-4(5)-carboxylate (**4**) needed for this reaction, was prepared as reported earlier.[1] Treatment of **4** with hydrazine hydrate in ethanol at reflux temperature gave an excellent yield of 5-amino-6-hydrazino-1,5-dihydroimidazo[4,5-*c*]pyridin-4-one (**5**) (Scheme I), which, on hydrogenation in the presence of Raney nickel (W-4) catalyst, provided **1** in 80% yield, which is a considerable improvement over a previously reported procedure[15] (52%). The advantage of this procedure over that previously reported[1,15] is that **1** could be readily obtained in good yield without the use of a high-pressure reaction vessel. A similar treatment of methyl 5-(cyanomethyl)-1-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)imidazole-4-carboxylate (**6**)[1] with hydrazine hydrate gave crystalline 1-amino-3-deazaguanosine (**8**). Hydrogenation of **8** in the presence of Raney nickel (W-4) catalyst provided a new synthesis of 3-deazaguanosine (**2**), which was obtained in good yield.

A plausible mechanism for the ring closure of **6** to form **8** may be visualized simply as occurring by the direct attack of hydrazine on the ester carbonyl carbon to give the intermediate **7**, which cyclizes immediately to form **8**. However, in the case of **4** (mechanism A), abstraction of the ring NH proton by hydrazine gives the anion **9**, which

Mechanism A



9 10 11 5

is followed by addition of hydrazine across the nitrile bond to give the intermediate **10**. Further nucleophilic attack of hydrazine on the ester carbonyl carbon gives the imidazole precursor **11**, which on ring closure and aromatization yields **5**.

For the synthesis of 3-deazaguanosine 3′,5′-cyclic phosphate (**13**) (Scheme II), 5-(cyanomethyl)-1-β-D-ribofuranosylimidazole-4-carboxamide 5′-phosphate[1] was found to be a versatile starting material. Treatment of the *N*,*N*′-dicyclohexylmorpholinecarboxamidine salt of 5-(cyanomethyl)-1-β-D-ribofuranosylimidazole-4-carboxamide 5′-phosphate in anhydrous pyridine with dicyclohexylcarbodiimide (DCC) according to the general procedure of Smith et al.[16] gave 5-(cyanomethyl)-1-β-D-ribofuranosylimidazole-4-carboxamide 3′,5′-cyclic phosphate (**12**). The appearence of a singlet for the anomeric proton in the ^{1}H NMR spectrum of **12** at δ 5.76 ppm indicated the formation of the cyclic phosphate ring. Ring closure of **12** in the presence of aqueous Na_2CO_3 provided **13**, which was isolated as the free acid.[17] Direct DCC-mediated cyclization of 3-deazaguanylic acid (**3**)[1] to obtain **13**, under the conditions of Smith et al.,[16] provided a complex reaction mixture from which the isolation of pure **13** was very difficult.

The antitumor mode of action of 3-deazaguanine has been postulated to be due to the consequence of its incorporation into tumor cell DNA.[5,9] To evaluate this hypothesis we undertook the synthesis of the 2′-deoxy derivatives of 3-deazaguanosine (**26–29**). While this work was in progress, a paper appeared[18] in which the syntheses of **26–29** were described. However, the physicochemical data reported for these compounds differ from the data we observed. As depicted in Scheme III, compound **4** was again proved to be a useful starting material. Silylation of **4** with hexamethyldisilazane gave the silylated product **14**. Reaction of **14** with 1 molar equiv of 1-*O*-methyl-2-deoxy-3,5-di-*O*-*p*-toluoyl-D-ribofuranose (**15**)[19] in CH_3CN in the presence of 1.44 molar equiv of trimethylsilyl trifluoromethanesulfonate (TMS-triflate) according to the procedure of Vorbrüggen et al.[20] gave a complex reaction

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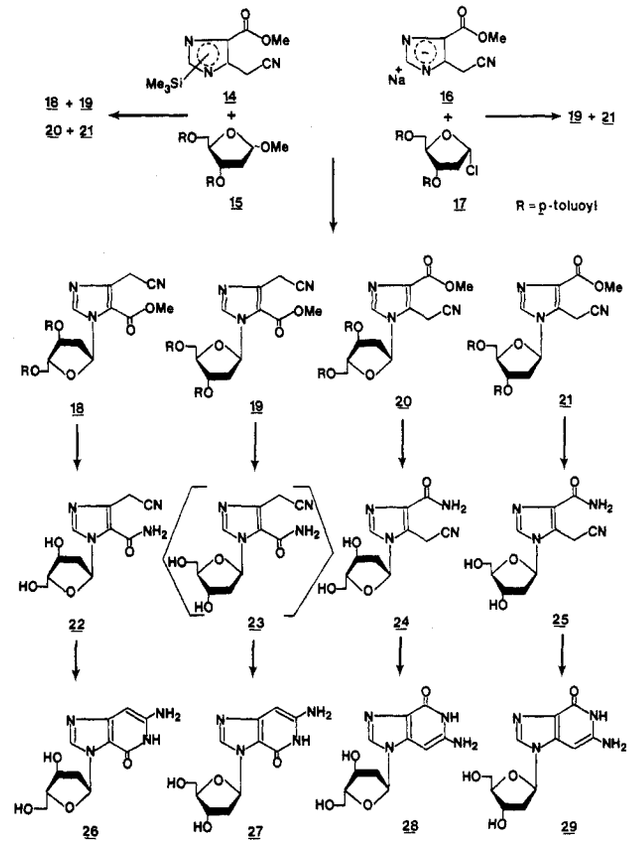
(20) Vorbrüggen, H.; Krolikiewicz, K.; Bennua, B. *Chem. Ber.* **1981**, *114*, 1234.

mixture of two major and two minor nucleosidic products along with trace amounts of unreacted sugar and the heterocycle. The separation of these products was achieved on a silica gel column by preparative LC techniques using acetone:hexane (1:4, v/v) as the solvent. The two major products were isolated as pure crystalline compounds and were identified as methyl 4-(cyanomethyl)-1-(2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentofuranosyl)imidazole-5-carboxylate (**18**, 20% yield) and methyl 5-(cyanomethyl)-1-(2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentofuranosyl)imidazole-4-carboxylate (**20**, 22% yield). The corresponding β-anomers **19** and **21** were isolated in less than 7% yield. The anomeric assignment of these isomers was determined by ^{1}H NMR wherein the β-isomers (**19** and **21**) exhibited the characteristic triplet for the anomeric proton and the α-isomers (**18** and **20**) the quartet[21] (see the Experimental Section). Although deoxyribosylation had occurred with equal facility at both imidazole ring nitrogens, this reaction condition favored the formation of α-anomers **18** and **20**. It has been well documented that the anomeric ratio of a Lewis acid mediated condensation of a silylated aglycon with a blocked 2-deoxy-D-*erythro*-pentofuranose is influenced by a number of factors, such as temperature, solvent, blocking groups, etc.[22-24] The apparent preponderance of the α-anomers in the present study seems to be influenced by the participation of the cyclic glycon acyloxonium ion involving 5′-*O*-toluoyl group.[25,26]

In an effort to improve the yield of the desired β-anomers **19** and **21**, the sodium salt glycosylation procedure, developed recently in our laboratory,[27-29] was employed. Application of this simple single-phase glycosylation procedure to the synthesis of **19** and **21** was found to be remarkably successful. Glycosylation of the sodium salt of **4** (**16**, generated in situ by the treatment of NaH in CH_3CN) with 1-chloro-2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentofuranose[30] (**17**) at ambient temperature gave a mixture of two nucleoside products (Scheme III). These products were separated on a silica gel column and identified as **19** (45% yield) and **21** (48.3% yield) on the basis of ^{1}H NMR characteristics (triplets for the anomeric protons). No formation of α-anomers was detected. Since the starting halosugar **17** has the α-configuration[31] in the solid state, the exclusive formation of the blocked 2′-deoxy-β-nucleosides in the present study is believed to be due to a direct Walden inversion (S_N2) at the C_1 carbon by the anionic heterocyclic nitrogen.

Subsequent treatment of each of the blocked imidazole nucleosides with liquid NH_3 provided the corresponding 7- and 9-substituted 3-deazaguanine 2′-deoxyribonucleosides. Thus, treatment of **18** with liquid NH_3 at 100 °C for 10 h, followed by silica gel column chromatography of the reaction residue, gave 6-amino-3,5-dihydro-3-(2-deoxy-α-D-*erythro*-pentofuranosyl)imidazo[4,5-*c*]pyridin-4-one (**26**) as a crystalline solid in a 69.5% yield, along with a minor amount of uncyclized 4-(cyanomethyl)-1-(2-deoxy-α-D-*erythro*-pentofuranosyl)imidazole-5-carboxamide (**22**). Similarly, treatment of **19** with CH_3OH/NH_3 at 100 °C for 18 h gave exclusively the cyclized product 6-amino-3,5-dihydro-3-(2-deoxy-β-D-*erythro*-pentofuranosyl)imidazo[4,5-*c*]pyridin-4-one (**27**). The ^{1}H NMR signal for the anomeric proton of **26** and **27** followed the "quartet–triplet" splitting pattern[21] for the α- and β-anomers, respectively. The observed peak width of the anomeric proton of 10.2 and 13.0 Hz for **26** and **27** was also consistent with the usual α and β values.[21] The UV absorption spectra of both **26** and **27** are virtually identical with that of 6-amino-3,5-dihydro-3-β-D-ribofuranosylimidazo[4,5-*c*]pyridin-4-one.[1]

Scheme III



When the blocked N-1 glycosyl derivatives of the imidazole (**20** and **21**) were treated with liquid NH_3 at 100 °C in a pressure vessel, crystalline 5-(cyanomethyl)-1-(2-deoxy-α-D-*erythro*-pentofuranosyl)imidazole-4-carboxamide (**24**) and the corresponding β-anomer **25** were obtained in 75.0% and 67.7% yield, respectively. It is of particular interest that the N-3 glycosyl isomers **18** and **19**, under similar reaction conditions, are mostly converted to **26** and **27**, which is in analogy to the direct cyclization of methyl 4-(cyanomethyl)-1-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)imidazole-5-carboxylate to 7-ribosyl-3-deazaguanine.[1] Base-catalyzed cyclization of **24** and **25** proceeded smoothly to yield 6-amino-1,5-dihydro-1-(2-deoxy-α-D-*erythro*-pentofuranosyl)imidazo[4,5-*c*]pyridin-4-one (**28**) and 2′-deoxy-3-deazaguanosine (**29**), respectively. Unlike the previous report,[18] both **28** and **29** were crystallized from water as needles. The UV absorption spectra of **28** and **29** [λ_{max} (pH 1) 283 and 310 (sh) nm; λ_{max} (pH 7) 269 and 295 (sh) nm; λ_{max} (pH 11) 271 nm] are virtually identical with that of 3-deazaguanosine.[1] The ^{1}H NMR spectrum of **29** in Me_2SO-d_6 displayed a pseudo-

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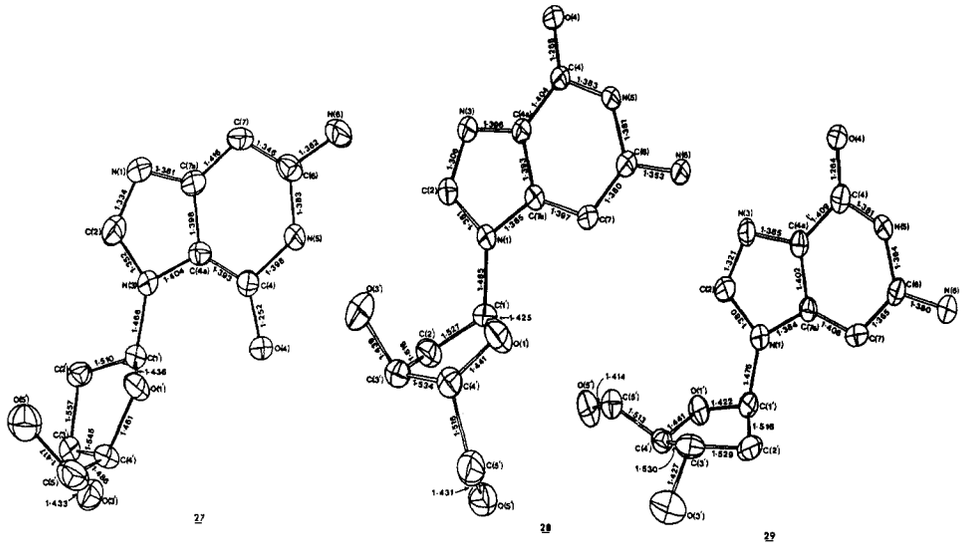

Figure 1. ORTEP drawing with the atom labels and bond lengths of compounds **27–29**.

triplet for the anomeric proton centered at δ 5.94 ppm (peak width 13.5 Hz), indicating the β-configuration. Similarly, compound **28** revealed a quartet for the anomeric proton centered at δ 5.95 (peak width 10.5 Hz), indicating the α-configuration. The observed 0.73 and 0.68 ppm dowfield shifts of the anomeric proton of **26** and **27** as compared to those of **28** and **29** can be attributed to the anisotropic effect of the carbonyl group on the anomeric proton in **26** and **27**, respectively, thereby providing additional support for the assigned site of glycosylation.

Since there were several discrepancies between our observed melting point data and that reported[18] for the same compounds (e.g., observed mp for **18**, 146–147 °C (lit. mp 133–135 °C); **19**, 132–133 °C (lit. mp 144–147 °C); **21**, 82–83 °C (lit. mp 60–62 °C); **25**, 156–158 °C (lit. mp 164–167 °C); **24**, 169–170 °C (not isolated); **27**, 147–149 °C (lit. mp >250 °C); **29**, 230–231 °C (lit. mp >250 °C), no melting point and elemental analysis for **26**; and no elemental analysis for **28**, etc.], it was deemed desirable to unequivocally substantiate our structural assignments by single-crystal X-ray analyses for 2′-deoxy-3-deazaguanosine (**29**), the α-anomer **28**, and 7-(2-deoxy-β-D-*erythro*-pentofuranosyl)-3-deazaguanine (**27**).

It would appear that the assignment of the anomeric configuration of the N-3 glycosylated imidazoles **18** and **19** has been erroneously reversed by Mian and Khwaja.[18] The melting point of our β-blocked nucleoside **19** matches that of the α-anomer described by Mian and Khwaja.[18] Similarly, the melting point for our α-anomer **18** corresponds to that of the β-anomer assigned by these authors.[18] These data also suggest the erroneous assignments may have been carried over to the ring-closed 7-(2-deoxy-D-ribofuranosyl)-3-deazaguanines **26** and **27**. Since the melting point and the chemical shift (in Me_2SO-d_6) of the anomeric proton in ^1H NMR spectrum of our α-anomer **26** correspond to those reported for the β-anomer of Mian and Khwaja,[18] we confirmed our structural assignment of the β-anomer **27** by single-crystal X-ray analysis. Unfortunately, Mian and Khwaja[18] did not report either a melting point or elemental analysis for the corresponding α-anomer, **26**.

Single-Crystal X-ray Diffraction Analysis of 27–29

Slow crystallization of **27–29** from water gave X-ray quality crystals. Data for the determination of lattice parameters and for the structural studies were collected by utilizing a Nicolet P3 autodiffractometer using graphite monochromated Mo Kα radiation (λ = 0.710 69 Å). The lattice parameters for the three nucleosides under study were obtained by using a least-squares technique of 15 centered 2θ values and the space groups were determined by using systematic extinction data. Single-crystal X-ray data were collected by using θ–2θ scan technique with variable scan rates, and the crystal data are summarized in Table I.

Table I

	compd **27**	compd **28**	compd **29**
space group	$P2_12_12_1$	$P2_12_12_1$	$P2_1$
a	6.987 (1) Å	8.165 (8) Å	4.754 (1) Å
b	9.237 (2) Å	10.933 (9) Å	11.199 (3) Å
c	19.626 (5) Å	13.483 (8) Å	11.993 (3) Å
β	90°	90°	91.54°
Z	4	4	2
sin θ/λ limit	0.70	0.65	0.65
unique reflctns	1942	1554	1425
reflctns used in refinement	1483[a]	1399[a]	1274[b]

[a] Those reflections with $F < 4\sigma(F)$ were not used in refinement. [b] Those reflections with $F < 2\sigma(F)$ were not used in refinement.

The structure of **27** was solved by using a combination of Patterson and direct method techniques, while the structures of **28** and **29** were solved by using direct methods. All structures were refined by using a full-matrix least-squares procedure. The crystallographic calculations were done by using the SHELX-76 program package.[32] The final residual values are as follows: for **27**, R = 0.062, R_w = 0.034; for **28**, R = 0.042 and R_w = 0.032 (weights for both **27** and **28** were based on counting statistics), and for **29**, R = 0.058 (unit weights were used). A perspective view of the molecules, drawn with the aid of the ORTEP program,[33] is shown in Figure 1 along with numbering of the atoms and bond lengths (Å).

The results of this structure-determination study unequivocally confirmed the β-anomeric configuration and the site of glycosyl attachment as N-1 in **29**, whereas **28** has the α-anomeric configuration and the glycosylation site as N-1. However, compound **27** has β-anomeric configuration and the glycosyl attachment on N-3.

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Table II. Comparative in Vitro Antiviral Activity of Ribavirin and 3-Deazaguanine and Its Derivatives in Vero Cells

compound	toxic level[a]	virus ratings[b]			
		HSV1	VV	Para 3	VSV
3-deazaguanine (1)	2.2	1.40	1.55	0.98	1.55
5-amino-1,5-dihydro-6-hydrazinoimidazo[4,5-c]pyridin-4-one (5)	240.0	0.05	0.15	0.03	0
3-deazaguanosine (2)	4.3	1.40	1.50	1.18	1.50
1-amino-3-deazaguanosine (8)	450.0	0.25	1.25	0.30	0.45
3-deazaguanosine 3′,5′-cyclic phosphate (13)	5.3	1.02	1.15	0.62	1.00
7-(2-deoxy-α-D-ribofuranosyl)-3-deazaguanine (26)	400.0	0.30	1.18	0.22	0.35
7-(2-deoxy-β-D-ribofuranosyl)-3-deazaguanine (27)	none	0.80	1.42	0.92	1.06
9-(2-deoxy-α-D-ribofuranosyl)-3-deazaguanine (28)	none	1.00	1.65	0.70	0.80
2′-deoxy-3-deazaguanosine (29)	none	1.28	1.72	1.45	1.03
1-β-D-ribofuranosyl-1,2,4-triazole-3-carboxamide (ribavirin)	none	0.60	1.20	1.30	0.80

[a] In micrograms per milliliter. [b] The virus rating (VR) was determined by comparing CPE development in drug-treated cells (T) and virus control cells (C). The CPE value (0–4) assigned to T for each drug level was subtracted from that of C, and the differences (C − T) were totaled. If partial toxicity was evident at any drug level, the C − T of that level was divided by 2. The sum of all C − T values was then divided by 10 times the number of test cups used per drug level.

Table III. In Vitro Antitumor Activity of 3-Deazaguanine and Its Derivatives

compound	ID_{50}[a]	
	L1210	P388
3-deazaguanine (1)	6.2×10^{-5} M	39% inhibtn at 10^{-4} M
5-amino-1,5-dihydro-6-hydrazinoimidazo[4,5-c]pyridin-4-one (5)	3.5×10^{-5} M	6.2×10^{-5} M
3-deazaguanosine (2)	1.8×10^{-5} M	1.6×10^{-5} M
1-amino-3-deazaguanosine (8)	b	b
3-deazaguanosine 3′,5′-cyclic phosphate (13)	4.1×10^{-6} M	4.5×10^{-5} M
7-(2-deoxy-α-D-ribofuranosyl)-3-deazaguanine (26)	8.0×10^{-5} M	31% inhibtn at 10^{-5} M
7-(2-deoxy-β-D-ribofuranosyl)-3-deazaguanine (27)	b	b
9-(2-deoxy-α-D-ribofuranosyl)-3-deazaguanine (28)	45% inhibtn at 10^{-5} M	b
2′-deoxy-3-deazaguanosine (29)	2.0×10^{-5} M	4.2×10^{-5} M

[a] Inhibitory dose 50 (ID_{50}) is the concentration of the compound in the culture media that produced 50% inhibition of the tumor cell growth as compared to the untreated controls. [b] Inactive at 10^{-4} M.

Biological Studies

Antiviral. 3-Deazaguanine and most of its derivatives synthesized during this study were tested against herpes simplex type 1 (HSV1), vaccinia (VV), parainfluenza type 3 (Para 3), and vesicular stomatitis (VSV) viruses in vitro in parallel with ribavirin (Table II). 3-Deazaguanine (1), 3-deazaguanosine (2), and 2′-deoxy-3-deazaguanosine (29), all exhibited potent activity against all the above viruses. 1-Amino-3-deazaguanosine (8) and 7-(2-deoxy-β-D-ribofuranosyl)-3-deazaguanine (27) showed potent activity only against VV. However, compounds 13 and 28 were found to inhibit the growth of HSV1 and VV significantly, in vitro. Compound 13 also showed marked activity against VSV. All other compounds are devoid of significant antiviral activity against these viruses in vitro.

Antitumor. The 3-deazaguanine nucleosides and nucleotides synthesized during this study were also tested against L1210 and P388 leukemia in vitro (Table III). All the compounds except 8, 27, and 28 were found to be moderate inhibitors of the growth of leukemia L1210 in vitro. 3-Deazaguanosine (2), the cyclic phosphate 13, 5-amino-6-hydrazino-1,5-dihydroimidazo[4,5-c]pyridin-4-one (5), and 2′-deoxy-3-deazaguanosine (29) are also significantly active against P388 leukemia in vitro. The other compounds under study were inactive. Thus, only 3-deazaguanine (1), 3-deazaguanosine (2), 2′-deoxy-3-deazaguanosine (29), and 3-deazaguanosine 3′,5′-cyclic phosphate (13) exhibited a broad spectrum of antiviral and antitumor activity in vitro.

Experimental Section

Melting points were taken on a Thomas-Hoover capillary melting point apparatus and are uncorrected. Nuclear magnetic resonance (^{1}H NMR) spectra were determined at 89.6 MHz with a JEOL FX-90Q spectrometer. The chemical-shift values are expressed in δ values (parts per million) relative to tetramethylsilane as an internal standard. The presence of water as indicated by elemental analysis was verified by ^{1}H NMR. Infrared spectra (IR) were obtained on a Beckman Acculab 2 spectrophotometer and ultraviolet spectra (UV; sh = shoulder) were recorded on a Cary Model 15 spectrophotometer. Elemental analyses were performed by Galbraith Laboratories, Inc., Knoxville, TN, and Robertson Labs, Florham Park, NJ. Thin-layer chromatography (TLC) was run on silica gel 60 F-254 plates (EM Reagents). J.T. Baker silica gel (70–230 mesh) was used for column chromatography. Preparative liquid chromatography (LC) was run by utilizing the Waters Prep 500 LC system. All solvents used were reagent grade. Detection of components on TLC was by UV light and with 10% H_2SO_4 in MeOH spray followed by heating. Evaporations were carried out under reduced pressure with the bath temperature below 30 °C.

5-Amino-6-hydrazino-1,5-dihydroimidazo[4,5-c]pyridin-4-one (5). To a suspension of methyl 5(4)-(cyanomethyl)-imidazole-4(5)-carboxylate[1] (4; 4.95 g, 30 mmol) in absolute EtOH (40 mL) was added hydrazine hydrate (99–100%, 5.0 g, 100 mmol) and the mixture was heated under reflux with the exclusion of moisture. Within a few minutes a clear solution was obtained and after 1 h solid started precipitating out. Heating was continued for further 15 h, after which time the reaction mixture was cooled to room temperature. The crystalline solid was collected, washed with EtOH (5 × 25 mL), followed by ether (3 × 50 mL), and air-dried to yield 5.35 g of the title compound. Recrystallization from water using decolorizing carbon provided an analytical sample (5.0 g, 92.6%) as light yellow needles: mp >300 °C (after drying at 80 °C for 15 h); IR (KBr) ν 1560 ($NHNH_2$), 1660 (C=O), 3120–3320 (NH, NH_2) cm^{-1}; UV λ_{max} (pH 1) 271 nm (ε 7700), 292 (sh) (5400); UV λ_{max} (pH 7) 262 nm (ε 6900), 299 (4800); UV λ_{max} (pH 11) 262 nm (ε 7400), 298 (3800); ^{1}H NMR (Me_2SO-d_6) δ 4.27 (br s, 2, $NHNH_2$), 5.35 (s, 2, N_5 NH_2), 5.93 (s, 1, C_7 H), 7.12 (br s, 1, $NHNH_2$), 7.76 (s, 1, C_2 H); MS, *m/e* 181 (M^+). Anal. ($C_6H_8N_6O$·0.25 H_2O) C, H, N.

6-Amino-1,5-dihydroimidazo[4,5-c]pyridin-4-one (3-Deazaguanine, 1). To a solution of 5 (1.80 g, 10 mmol) in boiling water (100 mL) was added freshly prepared Raney nickel catalyst (W-4, wet weight ~5 g), and the mixture was heated under reflux for 30 min. An additional amount of the catalyst (~3 g) was added, and refluxing was continued further for 30 min. The mixture was filtered hot through a Celite pad to remove the catalyst, which was washed well with boiling water (5 × 25 mL).

The combined filtrate and washings were decolorized with charcoal and concentrated to ~25 mL. It was allowed to stand at room temperature overnight in the dark. The light yellow needles that separated were collected and dried (at 100 °C for 12 h) to yield 1.20 g (80%): mp >320 °C [lit.[1] mp >350 °C]; IR (KBr) ν 1665 (C=O), 2950–3400 (NH_2) cm^{-1}; UV λ_{max} (pH 1) 274 nm (ϵ 11 500), 312 (6500); UV λ_{max} (pH 7) 262 nm (ϵ 10 500), 298 (8100); UV λ_{max} (pH 11) 262 nm (ϵ 10 000), 298 (7800); ^1H NMR (Me_2SO-d_6) δ 5.55 (s, 1, C_7 H), 5.65 (s, 2, NH_2), 7.82 (s, 1, C_2 H), 10.70 (br s, 1, NH), 12.35 (br s, 1, NH). Anal. ($C_6H_6N_4O$) C, H, N.

This compound was identical in all respects with 3-deazaguanine previously reported from our laboratory.[1]

5,6-Diamino-1,5-dihydro-1-β-D-ribofuranosylimidazo[4,5-*c*]pyridin-4-one (1-Amino-3-deazaguanosine, 8). To a solution of methyl 5-(cyanomethyl)-1-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)imidazole-4-carboxylate[1] (**6**; 12.19 g, 20 mmol) in absolute EtOH (125 mL) was added hydrazine hydrate (99–100%, 10.0 g, 200 mmol) and the mixture was heated under reflux. Within 2 h solid started precipitating out. Heating was continued for further 20 h. After cooling (ice bath), the solid was collected, washed with EtOH (5 × 40 mL) followed by ether (2 × 50 mL), and air-dried to yield 4.80 g of off-white solid. Crystallization from water (charcoal) provided analytically pure **8** as white needles: yield 4.50 g (75.7%); mp 254–255 °C dec; IR (KBr) ν 1680 (C=O), 3100–3400 (OH, NH_2) cm^{-1}; UV λ_{max} (pH 1) 287 nm (ϵ 11 600), 312 (sh) (5600); UV λ_{max} (pH 7) 276 nm (ϵ 11 300), 297 (sh) (8900); UV λ_{max} (pH 11) 276 nm (ϵ 11 000), 297 (sh) (8500); ^1H NMR (Me_2SO-d_6) δ 5.35 (s, 2, N_5 NH_2), 5.45 (d, 1, J = 5 Hz, $C_{1'}$ H), 5.60 (s, 1, C_7 H), 6.20 (s, 2, C_6 NH_2), 7.90 (s, 1, C_2 H), and other sugar protons. Anal. ($C_{11}H_{15}N_5O_5$) C, H, N.

6-Amino-1,5-dihydro-1-β-D-ribofuranosylimidazo[4,5-*c*]pyridin-4-one (3-Deazaguanosine, 2). To a solution of **8** (1.48 g, 5 mmol) in boiling water (50 mL) was added Raney nickel catalyst (W-4, wet weight ~3 g) and the mixture was heated under reflux for 45 min. After removal of the catalyst, the filtrate was worked up as described for **1**, and 3-deazaguanosine was obtained as white crystals after crystallization from water: yield 1.03 g (73%); mp 256–257 °C dec (lit.[1] mp 255–257 °C); IR (KBr) ν 1665 (C=O), 2920–3500 (OH, NH_2) cm^{-1}; UV λ_{max} (pH 1) 284 nm (ϵ 13 600), 309 (sh) (6500); UV λ_{max} (pH 7) 270 nm (ϵ 10 000), 298 (8000); UV λ_{max} (pH 11) 272 nm (ϵ 10 000), 295 (sh) (8000); ^1H NMR (Me_2SO-d_6) δ 5.50 (d, 1, J = 5.5 Hz, $C_{1'}$ H), 5.53 (s, 1, C_7 H), 5.66 (s, 2, NH_2), 7.93 (s, 1, C_2 H), 10.48 (s, 1, NH), and other sugar protons. Anal. ($C_{11}H_{14}N_4O_5$) C, H, N.

5-(Cyanomethyl)-1-β-D-ribofuranosylimidazole-4-carboxamide 3',5'-Cyclic Phosphate (12). To 5-(cyanomethyl)-1-β-D-ribofuranosylimidazole-4-carboxamide 5'-phosphate ammonium salt[1] (3.96 g, 10 mmol) in anhydrous pyridine (150 mL) was added *N,N'*-dicyclohexylmorpholinecarboxamidine[34] (2.93 g, 10 mmol), and the resulting solution was evaporated several times with dry pyridine to an anhydrous syrup. The dry syrup was dissolved in pyridine (750 mL) and added dropwise (over 1-h period) through a reflux condenser into a refluxing anhydrous solution of dicyclohexylcarbodiimide (DCC, 10.30 g, 50 mmol) in dry pyridine (2 L). The resulting solution was refluxed for an additional 15 h before water (125 mL) was added dropwise. After allowing to stand at room temperature for another 15 h, the solution was evaporated to dryness. To the residue were added water (125 mL) and ethyl ether (100 mL). The suspension was stirred vigorously and then filtered. The aqueous phase was separated and extracted with ethyl ether (3 × 100 mL). The aqueous layer was passed through a Dowex 50-X8 resin column (Na^+ form, 100–200 mesh, 2.5 × 25 cm), and the eluent was evaporated to a syrup. Ethanol (100 mL) was added to the syrup, and the resulting mixture was kept at room temperature for 24 h. The mixture was filtered and the clear filtrate was allowed to stand at 0–5 °C for a week. The resulting precipitate (1.5 g) was collected and dissolved in water (~5 mL), and the product was precipitated by adding ethanol (50 mL). The light brown product was dissolved in water (~5 mL) and silica gel (5 g) was added. The suspension was evaporated to a powder. Coevaporation with EtOH (3 × 50 mL) gave a dry residue, which was added on top of a silica gel column (2.5 × 30 cm, packed in $CHCl_3$), and the title compound was eluted with $CHCl_3$:MeOH (3:2, v/v). Evaporation of the appropriate homogeneous fractions gave 1.0 g (26%) of **12**: mp >220 °C dec; IR (KBr) ν 1670 (C=O), 2220 (weak, CN), 3400 (OH) cm^{-1}; UV λ_{max} (pH 1) 218 nm (ϵ 7200); UV λ_{max} (pH 7) 233 nm (sh) (ϵ 7500); UV λ_{max} (pH 11) 235 nm (sh) (ϵ 9200); ^1H NMR (D_2O) δ 4.48 (s, 2, CH_2), 5.76 (s, 1, $C_{1'}$ H), 7.98 (s, 1, C_2 H). Anal. ($C_{11}H_{12}N_4PNaO_7 \cdot H_2O$) C, H, N, P.

6-Amino-1,5-dihydro-1-β-D-ribofuranosylimidazo[4,5-*c*]pyridin-4-one 3',5'-Cyclic Phosphate (3-Deazaguanosine 3',5'-Cyclic Phosphate, 13). A solution of **12** (0.77 g, 2 mmol) in water (30 mL) was adjusted to pH 10.0 with 10% aqueous Na_2CO_3 solution and heated under reflux for 1 h. The ligh brown solution was concentrated to ~15 mL and then adjusted to pH 6.5 with Dowex-50 (H^+) resin before it was placed on a column of Bio-Rad AG-1 X 8 (formate form, 100–200 mesh, 30 mL). The column was first washed with water (500 mL) and then with water to 0.1 M formic acid gradient (2 L each). The product appeared after about 1.5 L of gradient had passed through the column. The fractions containing the homogeneous product were pooled, evaporated to a small volume, freezed, and lyophilized to provide 0.40 g (50%) of **13** as a light brown powder: mp >200 °C dec; UV λ_{max} (pH 1) 285 nm (ϵ 13 500), 309 (sh) (8300); UV λ_{max} (pH 7) 270 nm (ϵ 11 300), 297 (sh) (8500); UV λ_{max} (pH 11) 270 nm (ϵ 10 300), 296 (sh) (8300); ^1H NMR (D_2O) δ 5.90 (s, 1, $C_{1'}$ H), 8.80 (s, 1, C_2 H). Anal. ($C_{11}H_{13}N_4PO_7 \cdot 2H_2O$) C, H, N, P.

Methyl 5-(Cyanomethyl)-1-(2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D- and -β-D-*erythro*-pentofuranosyl)imidazole-4-carboxylate (18 and 19) and Methyl 4-(Cyanomethyl)-1-(2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D- and -β-D-*erythro*-pentofuranosyl)-imidazole-5-carboxylate (20 and 21). Method A. A mixture of dry **4** (4.95 g, 30 mmol), hexamethyldisilazane (HMDS, 60 mL), and ammonium sulfate (0.10 g) was heated under reflux for 15 h with the exclusion of moisture. Excess HMDS was removed by distillation to provide the trimethylsilyl derivative **14** as a yellowish brown oil. To the solution of **14** in dry CH_3CN (200 mL) was added 1-*O*-methyl-2-deoxy-3,5-di-*O*-*p*-toluoyl-D-ribofuranose[19] (**15**; 11.53 g, 30 mmol), followed by TMS-triflate[35] (9.60 g, 43.2 mmol). The clear reaction mixture was stirred at ambient temperature for 45 h. TLC (silica gel, acetone:hexane, 1:4) of an aliquot (treated with EtOH) indicated almost complete conversion of the sugar and the heterocycle to a multiple of products. The reaction mixture was evaporated to dryness, and the residue was dissolved in $CHCl_3$ (250 mL) and poured into a 5% aqueous $NaHCO_3$ solution (500 mL). The organic layer was separated and washed with 5% $NaHCO_3$ solution (2 × 100 mL), followed by water (3 × 50 mL). The dried (Na_2SO_4) organic extracts were evaporated, and the residual syrup was purified on a silica gel column by preparative LC techniques using acetone:hexane (1:4, v/v) as the solvent. The following four nucleosides were isolated in the order listed. **Methyl 4-(cyanomethyl)-1-(2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentofuranosyl)imidazole-5-carboxylate (18)**: white needles (MeOH), 3.1 g (20%); mp 146–147 °C; IR (KBr) ν 1720 (C=O), 2250 (CN) cm^{-1}; UV λ_{max} (pH 1) 247 nm (ϵ 20 200); UV λ_{max} (MeOH) 247 nm (ϵ 19 500); UV λ_{max} (pH 11) 254 nm (ϵ 17 800); ^1H NMR ($CDCl_3$) δ 2.38, 2.44 (2 s, 6, 2 CH_3), 3.95 (s, 3, CO_2CH_3), 4.60 (s, 2, CH_2), 6.71 (q, 1, $C_{1'}$ H, peak width 10.5 Hz), 7.26 (m, 4, Ph), 7.52 (s, 1, C_2 H), 7.96 (m, 4, Ph), and other sugar protons. Anal. ($C_{28}H_{27}N_3O_7$) C, H, N.

Methyl 4-(cyanomethyl)-1-(2-deoxy-3,5-di-*O*-*p*-toluoyl-β-D-*erythro*-pentofuranosyl)imidazole-5-carboxylate (19) was obtained as white needles (MeOH): 1.0 g (6.5%); mp 132–133 °C; IR (KBr) ν 1715 (C=O), 2240 (CN) cm^{-1}; UV λ_{max} (pH 1) 247 nm (22 200); UV λ_{max} (MeOH) 247 nm (ϵ 19 600); UV λ_{max} (pH 11) 255 nm (ϵ 18 800); ^1H NMR ($CDCl_3$) δ 2.40, 2.44 (2 s, 6, 2 CH_3), 3.95 (s, 3, CO_2CH_3), 4.67 (s, 2, CH_2), 6.75 (t, 1, $C_{1'}$ H, peak width 13.5 Hz), 7.26 (m, 4, Ph), 7.84 (s, 1, C_2H), 8.02 (m, 4, Ph), and other sugar protons. Anal. ($C_{28}H_{27}N_3O_7$) C, H, N.

Methyl 5-(cyanomethyl)-1-(2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentofuranosyl)imidazole-4-carboxylate (20) was isolated as a crystalline (MeOH) solid: 3.4 g (22%); mp 63–65 °C; IR (KBr) ν 1715 (C=O), 2260 (CN) cm^{-1}; UV λ_{max} (pH 1) 243

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nm (ε 33 600); UV λ_{max} (pH 7) 243 nm (ε 36 000); UV λ_{max} (pH 11) 243 nm (ε 21 400); ^{1}H NMR ($CDCl_3$) δ 2.41 (s, 6, 2 CH_3), 3.92 (s, 3, CO_2CH_3), 4.62 (s, 2, CH_2), 6.32 (q, 1, $C_{1'}$ H, peak width 10.0 Hz), 7.24 (m, 4, Ph) 7.76 (s, 1, C_2 H), 7.92 (m, 4, Ph), and other sugar protons. Anal. ($C_{28}H_{27}N_3O_7 \cdot H_2O$) C, H, N.

Methyl 5-(cyanomethyl)-1-(2-deoxy-3,5-di-*O*-*p*-toluoyl-β-D-*erythro*-pentofuranosyl)imidazole-4-carboxylate (21) was isolated as an amorphous solid: 0.75 g (4.8%); mp 82–83 °C; IR (KBr) ν 1720 (C=O), 2240 (CN) cm^{-1}; UV λ_{max} (pH 1) 244 nm (ε 30 600); UV λ_{max} (pH 7 and 11) 243 nm (ε 22 000); ^{1}H NMR ($CDCl_3$) δ 2.40, 2.44 (2 s, 6, 2 CH_3), 3.91 (s, 3, CO_2CH_3), 4.64 (s, 2, CH_2), 6.22 (t, 1, $C_{1'}$ H, peak width 14.0 Hz), 7.25 (m, 4, Ph), 7.73 (s, 1, C_2 H), 7.90 (m, 4, Ph), and other sugar protons. Anal. ($C_{28}H_{27}N_3O_7$) C, H, N.

Method B. To a suspension of **4** (4.95 g, 30 mmol) in anhydrous CH_3CN (100 mL) was added NaH (50% in oil, 1.60 g, 33 mmol), and the mixture was stirred at ambient temperature in an atmosphere of dry argon for 30 min, during which time a clear solution of **16** was obtained. Finely powdered dry (over KOH) 1-chloro-2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentofuranose[30] (**17**; 12.83 g, 33 mmol) was added portionwise and the resulting mixture was stirred further for 3 h under argon. The reaction mixture was filtered and to the filtrate was added silica gel (~30 g). The mixture was evaporated to dryness. Coevaporation with benzene (5 × 75 mL) gave dry residue, which was placed on top of a silica gel column (5 × 55 cm), prepacked in benzene. The column was eluted first with benzene:ethyl acetate (4:1.5, v/v, 2.5 L), followed by benzene:ethyl acetate (4:2, v/v). After the initial elution of the unreacted sugar **17** (0.50 g), methyl 4-(cyanomethyl)-1-(2-deoxy-3,5-di-*O*-*p*-toluoyl-β-D-*erythro*-pentofuranosyl)imidazole-5-carboxylate (**19**; 7.0 g, 45.1%), followed by methyl 5-(cyanomethyl)-1-(2-deoxy-3,5-di-*O*-*p*-toluoyl-β-D-*erythro*-pentofuranosyl)imidazole-4-carboxylate (**21**; 7.5 g, 48.3%) was obtained. Compounds **19** and **21** were found to be identical with that obtained by method A.

6-Amino-3,5-dihydro-3-(2-deoxy-α-D-*erythro*-pentofuranosyl)imidazo[4,5-*c*]pyridin-4-one (26). A mixture of **18** (1.03 g, 2 mmol) and liquid NH_3 (20 mL) was placed in a steel bomb (50 mL). The bomb was three-quarters submerged in a steam bath and heated for 10 h. The NH_3 was allowed to evaporate at room temperature, and the residue was subjected to a vacuum overnight to remove the last traces of NH_3. The residue was extracted several times with boiling ether (5 × 100 mL) to remove *p*-toluamide. The ether-insoluble material was purified on a silica gel column (2 × 40 cm, packed in EtOAc), using EtOAc:H_2O:1-PrOH, 4:2:1, upper layer, as the eluent. A small amount of 4-(cyanomethyl)-1-(2-deoxy-α-D-*erythro*-pentofuranosyl)imidazole-5-carboxamide (**22**; 30 mg, mp 115 °C) was isolated from the initial fractions. The later homogeneous fractions were pooled and evaporated to dryness, and the residue was crystallized from water (charcoal) to provide **26** as colorless needles, which, on exposure to light and air, developed color: 0.37 g (69.5%); mp >245 °C dec; IR (KBr) ν 1660 (C=O), 3200–3330 (OH, NH_2) cm^{-1}; UV λ_{max} (pH 1) 276 nm (ε 8800), 317 (5200); UV λ_{max} (pH 7) 260 nm (ε 5800), 316 (5500); UV λ_{max} (pH 11) 258 nm (ε 5800), 316 (5200); ^{1}H NMR (Me_2SO-d_6) δ 5.30 (br s, 2, C_6 NH_2), 5.50 (s, 1, C_7 H), 6.68 (q, 1, $C_{1'}$ H, peak width 10.2 Hz), 8.20 (s, 1, C_2 H), and other sugar protons. Anal. ($C_{11}H_{14}N_4O_4 \cdot H_2O$) C, H, N.

6-Amino-3,5-dihydro-3-(2-deoxy-β-D-*erythro*-pentofuranosyl)imidazo[4,5-*c*]pyridin-4-one (27). A mixture of **19** (5.17 g, 10 mmol), liquid NH_3 (80 mL), and anhydrous MeOH (20 mL) was heated in a steel bomb at 100 °C for 18 h. MeOH/NH_3 was evaporated and the brown residue was purified on a silica gel column (3.5 × 70 cm) using $CHCl_3$:MeOH (95:15, v/v) as the solvent. Crystallization of the pure material from water gave 1.55 g (58.2%) of the title compound: mp 147–149 °C dec; IR (KBr) ν 1625, 1660 (C=O), 3250–3360 (OH, NH_2) cm^{-1}; UV λ_{max} (pH 1) 277 nm (ε 13 300), 317 (6500); UV λ_{max} (pH 7) 258 nm (ε 8100), 317 (8400); UV λ_{max} (pH 11) 258 nm (ε 8000), 316 (7800); ^{1}H NMR (Me_2SO-d_6) δ 5.38 (br s, 2, C_6 NH_2), 5.49 (s, 1, C_7 H), 6.62 (t, 1, $C_{1'}$ H, peak width 13.0 Hz), 8.22 (s, 1, C_2 H), 10.53 (br s, 1, N_5 H), and other sugar protons. Anal. ($C_{11}H_{14}N_4O_4$) C, H, N.

5-(Cyanomethyl)-1-(2-deoxy-α-D-*erythro*-pentofuranosyl)imidazole-4-carboxamide (24). Compound **20** (5.17 g, 10 mmol) and liquid NH_3 (100 mL) were placed in a steel bomb (250 mL). The bomb was three-quarters submerged in a steam bath and heated for 10 h. At this point the TLC (silica gel, EtOAc:H_2O:1-PrOH, 4:2:1, upper phase) indicated almost complete conversion of the starting material to **24** and a minor amount of **28**. The ammonia was allowed to evaporate at room temperature, and the residue was subjected to a vacuum overnight to remove the last traces of NH_3. The residue was extracted several times with boiling ether (5 × 100 mL, to remove *p*-toluamide) and the ether-insoluble material was dissolved in MeOH, adsorbed on silica gel (~5 g), and placed on top of a silica gel column (4 × 40 cm, packed in EtOAc). The column was eluted with EtOAc:H_2O:1-PrOH (4:2:1, upper phase) and the appropriate homogeneous fraction were pooled and evaporated to dryness. Crystallization of the residue from water gave white needles: 2.0 g (75%); mp 169–170 °C; IR (KBr) ν 1660 (C=O), 2240 (CN), 3170–3450 (OH) cm^{-1}; UV λ_{max} (pH 1) 217 nm (ε 10 500); UV λ_{max} (pH 7 and 11) 235 nm (ε 9700); ^{1}H NMR (Me_2SO-d_6) δ 4.10 (m, 2, $C_{2'}$ H_2) 4.45 (s, 2, CH_2), 6.10 (q, 1, $C_{1'}$ H, peak width 10.0 Hz), 7.24 and 7.42 (2 br s, 2, $CONH_2$), 8.00 (s, 1, C_2 H), and other sugar protons. Anal. ($C_{11}H_{14}N_4O_4$) C, H, N.

6-Amino-1,5-dihydro-1-(2-deoxy-α-D-*erythro*-pentofuranosyl)imidazo[4,5-*c*]pyridin-4-one (28). A mixture of **24** (2.66 g, 10 mmol), 5% aqueous Na_2CO_3 solution (29 mL), and EtOH (15 mL) was heated under gentle reflux with stirring for 30 min. Complete dissolution was obtained as reflux started. The slightly brown solution was filtered, neutralized with Dowex-50 (H^+) resin, and evaporated to dryness. The residue was dissolved in MeOH, adsorbed on silica gel (~10 g), and placed on top of a silica gel column (4 × 40 cm, packed in EtOAc). The column was eluted with EtOAc:H_2O:1-PrOH (4:2:1, upper phase), and the homogeneous fractions were pooled and evaporated to dryness. The residue was crystallized from water as off-white needles: 2.2 g (83%); mp >240 °C dec; IR (KBr) ν 1650, 1670 (C=O), 3150–3320 (OH, NH_2) cm^{-1}; UV λ_{max} (pH 1) 283 nm (ε 13 000), 310 (sh) (6900); UV λ_{max} (pH 7) 270 nm (ε 13 200), 298 (9800); UV λ_{max} (pH 11) 271 nm (ε 12 500), 297 (sh) (9300); ^{1}H NMR (Me_2SO-d_6) δ 5.48 (s, 1, C_7 H), 5.60 (br s, 2, C_6 NH_2), 5.95 (q, 1, $C_{1'}$ H, peak width 10.5 Hz), 7.89 (s, 1, C_2 H), and other sugar protons. Anal. ($C_{11}H_{14}N_4O_4$) C, H, N.

5-(Cyanomethyl)-1-(2-deoxy-β-D-*erythro*-pentofuranosyl)imidazole-4-carboxamide (25). A mixture of **21** (5.17 g, 10 mmol) and liquid NH_3 (100 mL) was heated for 20 h at 100 °C, as described for **24**. After the removal of NH_3, the residue was purified on a silica gel column (5 × 50 cm, packed in EtOAc) using EtOAc:H_2O:1-PrOH (4:2:1, upper layer) as the eluent. The appropriate homogeneous fractions were pooled and evaporated to dryness. Crystallization of the residue from water (charcoal) gave white needles: 1.80 g (67.7%); mp 156–158 °C; IR (KBr) ν 1660 (C=O), 2260 (CN), 3350 (OH) cm^{-1}; UV λ_{max} (pH 1) 214 nm (ε 13 300); UV λ_{max} (pH 7) 233 nm (ε 24 500); UV λ_{max} (pH 11) 230 nm (ε 14 900); ^{1}H NMR (Me_2SO-d_6) δ 4.35 (s, 2, CH_2), 6.12 (t, 1, $C_{1'}$ H, peak width 13.5 Hz), 7.32 and 7.50 (2 br s, 2, $CONH_2$), 8.10 (s, 1, C_2 H), and other sugar protons. Anal. ($C_{11}H_{14}N_4O_4$) C, H, N.

6-Amino-1-(2-deoxy-β-D-*erythro*-pentofuranosyl)-imidazo[4,5-*c*]pyridin-4(5*H*)-one (2′-Deoxy-3-deazaguanosine, 29). A mixture of **25** (2.66 g, 10 mmol), 5% aqueous Na_2CO_3 solution (28 mL), and EtOH (30 mL) was heated under gentle reflux for 30 min. The reaction mixture was worked up and purified as described for **28** to yield 1.73 g (65%). Crystallization from water gave an analytical sample: mp 230–231 °C; IR (KBr) ν 1625, 1665 (C=O), 3220–3420 (OH, NH_2) cm^{-1}; UV λ_{max} (pH 1) 283 nm (ε 13 000), 310 (sh) (6700); UV λ_{max} (pH 7) 269 nm (ε 11 700), 295 (sh) (8500); UV λ_{max} (pH 11) 275 nm (ε 12 600); ^{1}H NMR (Me_2SO-d_6) δ 5.44 (s, 1, C_7 H), 5.59 (br s, 2, C_6 NH_2), 5.94 (t, 1, $C_{1'}$ H, peak width 13.5 Hz), 7.86 (s, 1, C_2 H), 10.32 (br s, 1, N_5 H), and other sugar protons. Anal. ($C_{11}H_{14}N_4O_4$) C, H, N.

Antiviral Evaluation. Inhibition of the virus-induced cytopathic effect (CPE) was used as the initial indicator of antiviral activity. CPE was observed in African green monkey kidney (Vero, V) cells after infection with herpes simplex type 1 (HSV 1, KOS), vaccinia (VV), parainfluenza type 3 (Para 3), and vesicular stomatitis (VSV) viruses. In this system, monolayers (18 h) of cells were exposed to the following $TCID_{50}$ (tissue culture 50% infective

dose) units of virus: HSV 1 (63), VV (200), Para 3 (56), VSV (3), and concentrations of each compound in one-half log dilutions ranging from 1000 to 1 μg/mL were added within 15–30 min. The degree of CPE inhibition was observed microscopically after 72 h of incubation at 37 °C in 5% CO_2 and scored numerically in order to calculate a virus rating (VR) as previously reported.[36] Significance of antiviral activity in terms of VRs has been assigned as follows: 0.5, slight or no activity; 0.5–0.9, moderate activity; ≥1.0, marked activity.

Antitumor Evaluation. L1210 leukemia and P388 lymphoid neoplasm were maintained in RPMI-1640 medium supplemented with 10% heat-inactivated fetal calf serum. For determination of cell growth inhibition, L1210 and P388 cells were seeded in 13 × 100 tubes at 5 × 10^4 cells/mL (2 mL/tube). Cells were grown in the presence of the compound of interest, at 4–5 log doses, for 48 h at 37 °C. Cell growth was assessed by cell count, using a Coulter cell counter. Cell growth at each dose level was expressed as a percentage of growth in control tubes and dose resulting in 50% inhibition of growth was determined.

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(1,3-Dialkyl-5-amino-1*H*-pyrazol-4-yl)arylmethanones. A Series of Novel Central Nervous System Depressants

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A series of novel (1,3-dialkyl-5-amino-1*H*-pyrazol-4-yl)arylmethanones was synthesized. Pharmacological evaluation of these compounds demonstrated central nervous sytem depressant activity, potential anticonvulsant properties, and a low order of acute toxicity. In addition, selected compounds showed potential antipsychotic effects. This report focuses on the synthesis and structure–activity relationships of these compounds. (5-Amino-1-ethyl-3-methyl-1*H*-pyrazol-4-yl)(2-chlorophenyl)methanone (**21**) was the most active compound against pentylenetetrazole-induced convulsions. (5-Amino-1,3-dimethyl-1*H*-pyrazol-4-yl)(3-chlorophenyl)methanone (**4**) also has a favorable anticonvulsant depression ratio. (5-Amino-1,3-dimethyl-1*H*-pyrazol-4-yl)(3-trifluoromethylphenyl)methanone (**8**), (5-amino-1,3-dimethyl-1*H*-pyrazol-4-yl)(3-thienyl)methanone (**13**), and (5-amino-3-ethyl-1-methyl-1*H*-pyrazol-4-yl)phenylmethanone (**14**) are very potent depressants. (5-Amino-1,3-dimethyl-1*H*-pyrazol-4-yl)(2-thienyl)methanone (**12**) possessed marked central depressant activity without anticonvulsant activity and without impairment of motor functioning. (5-Amino-1,3-dimethyl-1*H*-pyrazol-4-yl)(2-fluorophenyl)methanone (**2**) has a behavioral profile suggestive of antipsychotic activity and gave a positive Ames test result.

Epilepsy is a continuing medical problem despite the discovery and successful marketing of a number of drugs for its treatment;[1-3] thus the search continues for useful anticonvulsant agents.

A short series of novel (1,3-dialkyl-5-amino-1*H*-pyrazol-4-yl)arylmethanones had been prepared as intermediates in the synthesis of a series of compounds with antianxiety activity.[4,5] When these were submitted for pharmacological evaluation, some were found to possess interesting central nervous system depressant activity, potential anticonvulsant properties, plus a low order of acute toxicity. The series was expanded to attempt to maximize the level of activity and to study the structure–activity relationships. Encouraging activity was also observed in selected compounds when evaluated for potential antipsychotic activity.

Chemistry. Three general methods were developed for the synthesis of the title compounds. These are summarized in Chart I.

Route A. Many of the (1,3-dialkyl-5-amino-1*H*-pyrazol-4-yl)methanones were prepared by reaction of the (1,3-dialkyl-5-chloro-1*H*-pyrazol-4-yl)methanones[6] with concentrated ammonium hydroxide under heat and pressure.

Route B. A series of 1,3-dialkyl-5-amino-1*H*-pyrazoles were prepared by a variation of the method of Taylor and Hartke[7] in which 3-amino-2-butenenitrile was condensed with methylhydrazine. Larger alkylhydrazines do not ring close under neutral or basic conditions, the reaction stopping at the open alkylhydrazone stage. However, Koike et al.[8] used hydrogen chloride to effect this type of ring closure with phenylhydrazine. In our case, the addition of more than 1 equiv of concentrated hydrochloric acid to the larger alkylhydrazones resulted in high yields

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