(5050); ^{1}H NMR δ 0.95 (3 H, d, 5'-Me), 2.04 (3 H, s, Ac), 2.07 (3 H, s, Ac), 4.03 (1 H, m, H-3'), 4.38 (1 H, m, H-5'), 5.12 (1 H, dd, H-4', $J_{3',4'}$ = 3.7, $J_{4',5'}$ = 7.3 Hz), 5.17 (1 H, t, H-2', $J_{1',2'}$ = $J_{2',3'}$ = 3.2 Hz), 5.57 (1 H, dd, H-1', $J_{1',2'}$ = 3.2, $J_{1',3'}$ = 1.5 Hz), 5.69 (1 H, d, H-5, $J_{5,6}$ = 7.6 Hz), 7.56 (1 H, d, H-6), 8.64 (1 H, br s, NH).

Anal. Calcd for $C_{14}H_{17}N_3O_6 \cdot H_2O$: C, 49.26; H, 5.61; N, 12.31. Found: C, 49.53; H, 5.54; N, 12.44.

The HPLC analysis of the mother liquor of crystallization showed that there were two major components in the solution. Separation of these components was achieved by HPLC on a semipreparative reverse-phase column using H_2O–MeOH (70:30) as the mobile phase. From the first fraction (rV = 20 mL), after concentration in vacuo and crystallization of the residue from acetone, an additional 40 mg of **19** (total yield 47%) was obtained, mp 168–170 °C. The second fraction (rV = 29 mL) was concentrated and the residue crystallized from acetone. Compound **18** (80 mg, 42%) was obtained as colorless crystals: mp 172–174 °C; UV (MeOH) λ_{max} 262 nm (ε 7050), λ_{min} 230 (1520); ^{1}H NMR δ 1.10 (3 H, d, 5'-Me), 1.83 (3 H, s, NAc), 2.03 (3 H, s, OAc), 2.15 (3 H, s, OAc), 4.28 (1 H, m, H-5'), 4.68 (1 H, m, H-3'), 4.89 (1 H, br s, H-4'), 5.00 (1 H, br s, H-2'), 5.63 (1 H, d, H-5, $J_{5,6}$ = 8.1 Hz), 5.92 (1 H, s, H-1', $J_{1',2'}$ = 0 Hz), 7.45 (1 H, d, H-6), 7.76 (1 H, br d, N*H*Ac), 11.39 (1 H, br s, NH).

Anal. Calcd for $C_{16}H_{21}N_3O_8 \cdot {}^1/_3H_2O$: C, 49.35; H, 5.61; N, 10.79. Found: C, 49.43; H, 5.91; N, 10.66.

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Registry No. **1**, 32254-37-8; **2**, 32254-39-0; **3**, 96745-79-8; **4**, 80646-93-1; **5**, 96745-80-1; **6**, 96758-74-6; **7**, 96758-75-7; **8**, 96745-82-3; **9**, 96745-83-4; **10**, 14488-28-9; **11**, 96790-47-5; **12**, 96745-84-5; **13**, 4338-36-7; **14**, 96758-76-8; **15**, 96745-85-6; **16**, 96745-86-7; **17**, 32254-30-1; **18**, 96745-87-8; **19**, 96745-88-9.

Nucleic Acid Related Compounds. 48. Photoaddition of Methanol to 9-(β-D-Ribofuranosyl)purine (Nebularine) To Give Inhibitors of Adenosine Deaminase[1]

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Photolysis of anaerobic solutions of 9-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)purine (2',3',5'-tri-*O*-acetylnebularine) (**1b**) in methanol at 2537 Å gave a diastereomeric mixture of 9-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)-1,6-dihydro-6(*R*,*S*)-(hydroxymethyl)purines (**2b**) plus secondary photoproducts. The presence of oxygen resulted in more rapid formation of the 6-(hydroxymethyl)- (**3b**), 1,6-dihydro-6,6-bis(hydroxymethyl)- (**4b**), 6-methyl- (**5b**), and (*R*,*S*)-1,6-dihydro-6-(hydroxymethyl)-6-methyl-9-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)purine (**6b**) byproducts. Reevaluation of recently published claims that photoaddition of methanol to nebularine proceeded with high stereoselectivity is presented based on definitive ^{1}H and ^{13}C NMR spectral data and FAB mass spectrometry.

Adenosine deaminase (adenosine aminohydrolase EC 3.5.4.4) is a crucial catabolic enzyme in the regulation of metabolism of adenosine and 2'-deoxyadenosine.[2] It also effects deamination (and related hydrolytic displacements) of a number of adenine nucleoside analogues and thereby diminishes their effectiveness as drugs.[3] There has been considerable recent interest in the pharmacological evaluation of inhibitors of this enzyme as single agents and as substrate drug/inhibitor combinations.[4]

The naturally occurring 3-(β-D-ribofuranosyl)- (coformycin, Ia)[5] and 3-(2-deoxy-β-D-*erythro*-pentofuranosyl)-3,6,7,8-tetrahydroimidazo[4,5-*d*][1,3]diazepin-8(*R*)-ol (2'-deoxycoformycin, covidarabine, Ib)[6] nucleosides are the most potent known inhibitors of adenosine deaminase.[7,8] These two compounds are thought to function as "transition-state analogue" inhibitors.[9-11] Photoaddition of methanol to 9-(β-D-ribofuranosyl)purine (nebularine) (**1a**) was reported to give a product, 1,6-dihydro-6(hydroxymethyl)nebularine (**2a**),[12] that is a strong reversible inhibitor of this enzyme.[8,9,11,12] It also is thought to function as a transition-state analogue[9,11-13] with a less complementary fit at the catalytic center than I.

Our continuing interest in substrate binding to adenosine deaminase[3b,14] led us to consider evaluation of related putative tight-binding nucleoside analogues. Some of our prior results were not in harmony with conclusions pub-

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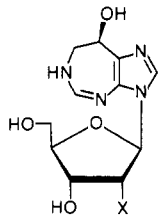
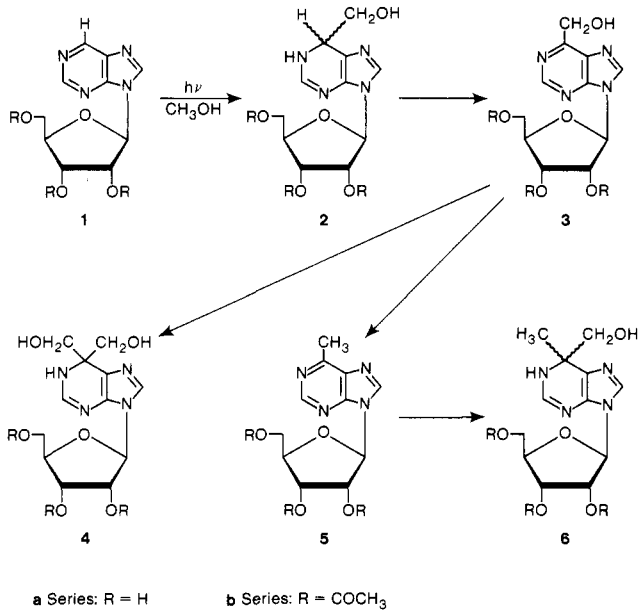


I a X = OH
b X = H

lished very recently[15] on the course of photoaddition of methanol to nebularine.

Photoaddition of alcohols to the parent purine ring was studied by Linschitz and Connolly.[16] They observed attachment of hydrogen at N1 and α-hydroxyalkyl at C6 under anaerobic photolysis. Evans and Wolfenden employed photoaddition of methanol to nebularine (**1a**) and reported chromatographic isolation of three products.[12] The two slower migrating products had ultraviolet absorption maxima at ~292 nm and gave mass spectral "parent ions" at m/z 284.[12] These two compounds were thought to be the 6-diastereomers of 1,6-dihydro-6-(hydroxymethyl)nebularine (**2a**).[9,12] The third product had a UV absorption maximum at 263 nm and other spectral properties compatible with the oxidatively aromatized 6-(hydroxymethyl)nebularine (**3a**).[12]



a Series: R = H b Series: R = $COCH_3$

Ohno *et al.*[17] subsequently reported that photoaddition of methanol to 2′,3′,5′-tri-*O*-acetylnebularine (**1b**) gave a 1,6-dihydro-6-(hydroxymethyl) adduct (**2b**) in 96% yield. This adduct was considered to be "free of isomeric impurities"[17] and was used in the first reported synthesis of coformycin. The photoaddition/ring expansion sequence was used again by these authors in their more recently reported low-yielding synthesis of coformycin (diastereomeric at C8) and isocoformycin.[18]

Wolfenden and co-workers had amplified their investigation of the photoaddition of methanol to nebularine.[13] They reported selective formation of 1,6-dihydro-6-(hydroxymethyl)nebularine (**2a**) under nitrogen, but all three products noted earlier[12] were obtained in the presence of air.[13] The fastest migrating product (6-(hydroxymethyl)nebularine, **3a**) was isolated and resubjected to photolysis in methanol. A product was obtained that had the same chromatographic mobility and ^1H NMR spectrum as the slowest migrating product. This compound was reassigned the structure of 1,6-dihydro-6,6-bis(hydroxymethyl)nebularine (**4a**) (rather than a mono adduct diastereomer).[13] The product of anaerobic photoaddition migrated with the middle product of the three observed in the presence of air. Narrowly shifted twinning of some sugar and 6-hydroxymethyl carbon signals in the ^{13}C NMR spectrum of this product was compatible with formation of comigrating C6 diastereomers.[13]

In 1983 Ohno and co-workers published a detailed reinvestigation of these photoaddition reactions.[15] Nebularine (**1a**) was reported[15] to give the same three major products found originally by Evans and Wolfenden.[12] A crystal obtained by slow (over one month) diffusion–crystallization of the major middle-migrating product fraction was subjected to X-ray diffraction and found to be 1,6-dihydro-6(*S*)-(hydroxymethyl)-9-(β-D-ribofuranosyl)purine (**2a** *S* isomer).[15] The slowest migrating product then was assigned as the diastereomeric 1,6-dihydro-6(*R*)-(hydroxymethyl) mono adduct (**2a**, *R* isomer).[15]

These authors reported that photolysis of 2′,3′,5′-tri-*O*-acetylnebularine (**1b**) in methanol gave three analogous product bands. The middle and slowest migrating products were assigned as the 6(*S*) (major) and 6(*R*) (minor) mono hydroxymethyl adducts (**2b**). The fastest migrating product (**3b**) was converted chemically to the known 6-methylnebularine (**5a**). Chemical oxidation of the major product gave the aromatized 6-(hydroxymethyl)purine nucleoside derivative (**3b**) which further corroborated the site of addition as C6.[15]

Several groups have employed the "1,6-dihydro-6-(hydroxymethyl)-9-(β-D-ribofuranosyl)purine photoadduct" in studies on the inhibition of adenosine deaminase. The kinetic constants obtained have been attributed to the presumed diastereomeric mixture[8,11,13,19,20] or the assigned individual stereoisomers.[12,15] The most recently published study has further confused this issue.[15] The slowest migrating (TLC) product, originally thought[9,12] to be a mono adduct (**2a**) but later assigned as the bis(hydroxymethyl) compound (**4a**) by Wolfenden[13] was reassigned as the mono adduct *R* diastereomer by Ohno.[15] The possibility of selective fractional crystallization of the 6(*S*) mono adduct from a mixed diastereomeric middle fraction was not addressed.[15] We consider it useful to report our results which confirm the latter interpretation and the earlier conclusions of Wolfenden and co-workers.[13]

Results and Discussion

Photolysis of 2′,3′,5′-tri-*O*-acetylnebularine (**1b**) in methanol under argon resulted in formation of a major (middle) and two minor product bands (TLC). With careful deoxygenation,[21] about 75% yields of the mono adduct (**2b**) (middle fraction) were obtained with very minor amounts of starting **1b** and the rapidly migrating

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(21) In contrast to the results reported in ref 15, we observed little or no difference in conducting the photolyses in quartz or Vycor vessels. However, a marked effect of oxygen on product formation and generation of secondary photoproducts was observed. Argon was bubbled through the reaction solutions for at least an hour before and continuously during the "anaerobic photolyses".

product band. Photolysis and sequential monitoring (TLC) of a less rigorously deoxygenated[21] solution of **1b** in methanol indicated initial formation of the major (middle) product (**2b**), followed by appearance of the fastest and then slowest product bands.

A sample of the fastest band (incompletely separated from the middle fraction by flash chromatography[22]) was found to contain several compounds by high-field ^{1}H NMR analysis. The major component in this sample was the mono hydroxymethyl 1,6-dihydro adduct **2b** (middle fraction overlap). The oxidatively aromatized 6-(hydroxymethyl)purine nucleoside derivative (**3b**), its deoxygenated[23] 6-methylpurine product (**5b**), and the 6-(hydroxymethyl)-6-methyl-1,6-dihydro photoadduct (**6b**) of the latter also were present.

Photolysis of a methanolic solution of **1b** without deoxygenation (and open to the atmosphere) resulted in disappearance of starting material and isolation of a 19% yield of 6-methyl-9-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)-purine (**5b**) after column chromatography of the intensely yellow reaction mixture. This product was identical with that prepared from the 6-(hydroxymethyl)purine compound (**3b**) by the published[15] chemical sequence.

Anaerobic photolysis of a methanolic solution of the 6-methylpurine derivative (**5b**) gave the 1,6-dihydro-6-(hydroxymethyl)-6-methylpurine nucleoside adduct (**6b**). The 400-MHz ^{1}H NMR spectrum of **6b** in $CDCl_3$ had narrowly shifted splitting of the base and certain resolved sugar proton peaks resulting from an essentially equal mixture of C6 diastereomers. Narrowly shifted twinning of a number of ^{13}C NMR signals also was observed.

Photolysis of a methanolic solution of the 6-(hydroxymethyl)purine compound (**3b**) (prepared from **2b** by chemical oxidation[15]) gave the 1,6-dihydro-6,6-bis(hydroxymethyl)purine adduct (**4b**). This compound had TLC mobility and spectral properties identical with those of the slowest migrating product from photolysis of **1b**. The ^{13}C NMR chemical shifts of **4b** in D_2O are in good agreement with those reported by Shimazaki et al. for their slowest migrating product.[15] However, it is apparent from comparison of the peak intensites in the spectra of **2b** and **4b** that C6 of **4b** is a quaternary carbon. There is no bonded-proton coupling and NOE effect for the signal from C6 of **4b** and the 6-CH_2OH carbon signal intensity is high for **4b**. There are differences noted in our 200-MHz ^{1}H NMR spectrum of **4b** (see the Experimental Section), but it is in general relative agreement with the one reported at 100 MHz.[15] No resonance was reported for H6 (the diastereomeric proton required in their structure) and a two-proton signal was noted at δ 4.15 for their 6-CH_2OD group.[15] Our spectrum has a narrowly shifted (0.002 ppm) doublet (of doublets) at δ 3.84 and a doublet at δ 3.61, each with $|J_{gem}| \sim 12$ Hz and integrating for two protons, for the four diastereotopic protons of the 6-CH_2OD groups of **4b**. This spectrum has no peak in the region of the signal for H6 in the spectrum of **2b**.

The reported EI mass spectrum of **4b** had the highest noted m/z ion at "452 $(M + Ac)^+$".[15] Our high resolution EI mass spectrum had m/z 482.1626 (0.3%, M − H + Ac, calcd for $C_{20}H_{26}N_4O_{10}$ 482.1649) and 451.1460 (9.1%, M − H + Ac − CH_2OH, calcd for $C_{19}H_{23}N_4O_9$ 451.1465). Our FAB mass spectrum of **4b** had m/z 441 (M + H). The EI mass spectrum of our 6-(hydroxymethyl)-6-methyl adduct (**6b**) also had no parent ion peak. The highest m/z ion for **6b** was 393.1410 (14%, M − CH_2OH, calcd for $C_{17}H_{21}N_4O_7$ 393.1410). However, the FAB mass spectrum of **6b** had peaks for positive ion-associated molecular species at m/z 425 (M + H), 447 (M + Na), and 463 (M + K).

^{1}H NMR spectra of the major (middle fraction) photoadduct (**2b**) at 100 MHz in D_2O had sharp individual peaks as reported by Shimazaki et al.[15] However, at 400 MHz (and with better resolution in $CDCl_3$ solutions) the diastereomeric complexity was apparent. At 100.6 MHz the ^{13}C NMR spectra of **2b** in both D_2O and $CDCl_3$ had narrowly resolved shifts for a number of carbons of the diastereomers.

Conclusions

The results obtained in these investigative experiments demonstrated that photolysis of methanolic solutions of 2′,3′,5′-tri-*O*-acetylnebularine (**1b**) resulted in a more complex reaction profile than reported earlier. It also is clear that *no significant stereoselectivity* occurred in the formation of the 1,6-dihydro-6-(hydroxymethyl) (**2b**) or 1,6-dihydro-6-(hydroxymethyl)-6-methyl (**6b**) photo adducts. This is in harmony with the earlier results of Wolfenden and co-workers[13] but in sharp contrast with the recent claims of Shimazaki et al.[15] Confirmatory reexamination of our earlier experiments in view of their recently published study leads to the conclusion that their optimistic views on the stereoselective[15] photoaddition of methanol to nebularine resulted from an unfortunate combination of overlapping NMR chemical shifts in the monoadduct diastereomers, loss of a hydroxymethyl fragment in EI mass spectra of the bis adduct, and selective crystallization of a single diastereomer for X-ray analysis.

Experimental Section

Photolyses were performed in a double-walled quartz[21] tube with cooling at 2 °C (with a Lauda Brinkman bath and circulation pump RC 20) in a Rayonet reactor with 16 75-W RPR 2537 Å lamps. Flash evaporation was conducted at or below room temperature under water aspirator or mechanical oil pump vacuum. Merck silica on plastic sheets (no. 5735) was used for thin-layer chromatography (TLC) with MeOH/$CHCl_3$ (1:9) as developing solvent and migrating fractions were visualized under a 2537 Å lamp. Flash chromatography[22] was performed with Merck silica no. 9385, 40–63 μm, and open column chromatography with Merck silica no. 7734, 63–200 μm. Solvent A is MeOH/$CHCl_3$ (1:19) and B is MeOH/$CHCl_3$ (1:9). Reagent grade methanol was redistilled for the photolyses.

Preparation of 2′,3′,5′-Tri-*O*-acetylnebularine (1b). Acetylation of 25 g (93 mmol) of inosine was effected by stirring with 250 mg of 4-(*N*,*N*-dimethylamino)pyridine and 250 mL of Ac_2O at room temperature.[24] Evaporation of volatile materials and coevaporation with toluene gave a residue that was crystallized from MeOH/$CHCl_3$ to give 35.4 g (96%) of 2′,3′,5′-tri-*O*-acetylinosine, mp 240–241 °C (lit.[25] mp 241 °C); ^{1}H NMR and mass spectra were in agreement with this known structure.

Chlorination of this material with $SOCl_2$/$HCONMe_2$/$CHCl_3$ as reported[26] followed by column chromatography (A) gave 9-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)-6-chloropurine[26b] as a colorless oil with UV, ^{1}H NMR, and mass spectra as expected for this known product.

A sample (780 mg, 1.9 mmol) of this material was hydrogenolyzed[27] with 300 mg (3.7 mmol) of NaOAc and 50 mg of 5% Pd·C catalyst in 15 mL of dry MeOH at 40 psi H_2 for 24 h at room

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temperature in a Parr shaking apparatus. Filtration of the mixture with a Celite pad, evaporation of solvent, partitioning with Et-OAc/H_2O, drying the organic phase ($MgSO_4$), evaporation of solvent, column chromatography, and then flash chromatography (A) gave 560 mg (78%) of the title compound as a somewhat hygroscopic colorless solid foam. This product was TLC homogeneous in two systems and had a clean, sharply resolved ^1H NMR spectrum[15] and mass spectrum in agreement with the known structure.

Photolysis of 1b under Argon. A solution of 920 mg (2.4 mmol) of **1b** in 90 mL of MeOH was purged with argon for at least 1 h,[21] cooled to 2 °C, and irradiated for 140 min with constant argon purging. TLC indicated formation of a slower migrating major product band plus essentially equivalent intensity minor faster migrating spots corresponding to starting **1b** and a slightly slower migrating zone. Solvent was evaporated from the reaction mixture and the residue was subjected to flash chromatography (B). Evaporation of appropriately pooled slower migrating fractions gave 770 mg (77%) of **1,6-dihydro-6-(hydroxymethyl)-9-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)purine (2b)** as a slightly yellow solid foam: UV (MeOH) max (broad) 298 nm (ε 5040); ^1H NMR (400 MHz, $CDCl_3$) δ 7.40 (Δδ 0.01) (2s, 1, H8), 7.02 (Δδ 0.007) (d of d becomes 2 s with D_2O exchange, 1, H2), 6.38 (br s, 1, NH, disappears with D_2O exchange), 5.88 (Δδ 0.012) (d of d, $J_{1'-2'}$ ~5 Hz, 1, H1'), 5.78 (Δδ 0.012) d of t, $J_{2'-3'}$ ~5 Hz, 1, H2'), 5.58 (Δδ 0.004) (d of t, $J_{3'-4'}$ ~5 Hz, 1, H3'), 5.04 (m, 1, H6), 4.40 (m, 3, H4',5',5''), 3.88 (Δδ 0.007) (d of t, J ~3 and 11 Hz, 1) and 3.76 (m, 1) (6-CH_2OH), 2.14, 2.12, 2.11 (3 s, 9, OAc's). A less well resolved spectrum with most peaks shifted slightly downfield was obtained in D_2O. ^{13}C NMR (100.6 MHz, $CDCl_3$) δ 170.42, 169.52, 169.40 (CH_3CO's), 146.80 (C2), 131.84 + 131.70 (C8), 86.13 + 85.94 (C1'), 79.86 (C4'), 73.46 + 73.44 (C2'), 70.69 + 70.66 (C3'), 66.79 (6-CH_2OH), 63.26 (C5'), 55.69 + 55.66 (C6), 20.68, 20.43, 20.38 (CH_3CO's); ^{13}C NMR (100.6 MHz, D_2O) (differences in our spectrum from those reported[15] were verified by selective bonded-proton decoupling) δ 173.79, 172.88, 172.56 + 172.51 (CH_3CO's), 149.65 (C2), 135.26 + 135.19 (C4), 132.15 + 132.08 (C8), 117.33 + 117.27 (C5), 85.45 + 85.37 (C1'), 80.02 + 79.96 (C4'), 73.97 + 73.93 (C2'), 70.86 + 70.82 (C3'), 65.28 (6-CH_2OH), 63.49 + 63.43 (C5'), 54.80 (C6), 20.49, 20.26, 20.12 (CH_3CO's); EI mass spectrum (270 °C), m/z 452.1539 (1.7%, M − H + Ac, calcd for $C_{19}H_{24}N_4O_9$ 452.1543), 410.1454 (1.4%, M, calcd for $C_{17}H_{22}N_4O_8$ 410.1438), 393.1406 (2.4%, M − OH, calcd for $C_{17}H_{21}N_4O_7$ 393.1410), 379.1249 (18.5%, M − CH_2OH, calcd for $C_{16}H_{19}N_4O_7$ 379.1254).

Photolysis of 1b with "Some" Oxygen Present. A solution of 160 mg (0.42 mmol) of **1b** in 80 mL of MeOH was purged with argon for 10 min and then was photolyzed as described above (without Ar purging) with magnetic stirring. The reaction progress was monitored periodically by removal of a 100-μL sample, evaporation to dryness under a stream of N_2, and dissolving the residue in CH_2Cl_2 for evaluation by TLC. At 30 min essentially all of starting **1b** had disappeared. Monoadduct **2b** and a faster migrating faint spot were observed. At 90 min a slower moving product spot also was visible. After 310 min the reaction mixture was evaporated and the residue subjected to flash chromatography. Evaporation of combined early fractions gave 88 mg of an oily material whose 400 MHz NMR spectrum in $CDCl_3$ indicated the presence (in the approximate chemical yield) of monoadduct **2b** (35%), the aromatized 6-hydroxymethyl compound **3b** (13%), its deoxygenated[23] 6-methylnebularine derivative **5b** (0.3%), and the product (**6b**, 3%) of photoaddition of methanol to the latter compound.

A TLC homogeneous slow migrating fraction was pooled and evaporated to give 42 mg (23%) of **1,6-dihydro-6,6-bis(hydroxymethyl)-9-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)purine (4b)**: UV (MeOH) max (broad) 296 nm (ε 4710); ^1H NMR (200 MHz, D_2O) δ 7.74 (s, 1, H8), 7.34 (s, 1, H2), 6.07 (d, $J_{1'-2'}$ ~5 Hz, 1, H1'), 5.76 ("t", $J_{2'-3'}$ ~5.5 Hz, 1, H2'), 5.59 ("t", $J_{3'-4'}$ ~5.5 Hz, 1, H3'), 4.60 (m, 1, H4'), 4.40 (m, 2, H5',5''), 3.84 (2d, Δδ 0.002, J_{gem} ~−12 Hz, 2) and 3.61 (d, J_{gem} ~−12 Hz, 2) (two nonequivalent 6-CH_2OD groups), 2.22, 2.18, 2.16 (3 s, 9, OAc's); ^{13}C NMR (100.6 MHz, D_2O) δ 174.0, 173.0, 172.7 (CH_3CO's), 149.8 (C2), 136.5 (C4), 132.5 (C8), 118.1 (C5), 85.5 (C1'), 80.1 (C4'), 73.9 (C2'), 70.8 (C3'), 65.5 (6-CH_2OH's), 64.1 (C6), 63.5 (C5'), 20.5, 20.3, 20.1 (CH_3CO's); EI mass spectrum (220 °C), m/z 482.1626 (0.3%, M − H + Ac, calcd for $C_{20}H_{26}N_4O_{10}$ 482.1649), 451.1460 (9.1%, M − H + Ac − CH_2OH, calcd for $C_{19}H_{23}N_4O_9$ 451.1465), 409.1376 (9.8%, M − CH_2OH, calcd for $C_{17}H_{21}N_4O_8$ 409.1359); FAB Ms, m/z 441 (M + H).

Photolysis of 1b in the Presence of Oxygen. A solution of 380 mg (1 mmol) of **1b** in 85 mL of MeOH was stirred magnetically at 2 °C and photolyzed as described above while open to the atmosphere for 90 min. TLC indicated that all of the starting **1b** had disappeared. The intensely yellow solution was evaporated and the residue was subjected to column chromatography ($CHCl_3/Me_2CO$, 1:1). Evaporation of the TLC homogeneous faster migrating fractions gave 75 mg (19%) of **6-methyl-9-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)purine (5b)**: ^1H NMR (400 MHz, $CDCl_3$) δ 8.87 (s, 1, H8), 8.19 (s, 1, H2), 6.25 (d, $J_{1'-2'}$ ~5.5 Hz, 1, H1'), 5.98 ("t", $J_{2'-3'}$ ~5.5 Hz, 1, H2'), 5.70 (d of d, $J_{3'-4'}$ ~5 Hz, 1, H3'), 4.49 (m, 2, H4',5'), 4.40 (d of d, 1, H5''), 2.88 (s, 3, 6-CH_3), 2.17, 2.14, 2.10 (3s, 9, OAc's); EI mass spectrum (200 °C), m/z 392.1322 (0.7%, M, calcd for $C_{17}H_{20}N_4O_7$ 392.1332). This product had identical TLC migration and spectral properties with those of a sample prepared by the reported[15] chemical sequence **2b** → **3b** →→→ **5b**.

Photolysis of 3b under Argon. A sample of the dihydro monohyroxymethyl adduct (**2b**) was oxidized with phenyltrimethylammonium perbromide to give **3b** as described.[15] A solution of 250 mg (0.61 mmol) of **3b** in MeOH was purged with argon for 1 h[21] and irradiated for 150 min at 2 °C with constant argon purging. Evaporation and flash chromatography (B) of the residue gave 102 mg (38%) of TLC homogeneous 6,6-bis(hydroxymethyl) adduct (**4b**) in addition to faster migrating components including **5b** and **6b**. This sample of **4b** had TLC mobility and UV, ^1H and ^{13}C NMR, and mass spectral properties identical with those of the slowest moving product of photolysis of **1b**.

Photolysis of 5b under Argon. A solution of 255 mg (0.675 mmol) of **5b** in 85 mL of MeOH was purged with argon for 1 h,[21] cooled to 2 °C, and photolyzed as described above for 135 min with constant argon purging. Evaporation of the solution and flash chromatography (B) gave 146 mg (53%) of TLC homogeneous **1,6-dihydro-6-(hydroxymethyl)-6-methyl-9-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)purine (6b)** as an essentially equal diastereomeric mixture: UV (MeOH) max (broad) 294 nm (ε 4130); ^1H NMR (400 MHz, $CDCl_3$) δ 7.36 (Δδ 0.01) (2s, 1, H8), 7.02 (Δδ 0.005) (2 br s, 1, H2), 5.90 (Δδ 0.005) (2d, $J_{1'-2'}$ ~5 Hz, 1, H1'), 5.77 ("t", $J_{2'-3'}$ ~5.2 Hz, 1, H2'), 5.60 (Δδ 0.006) (2 t, $J_{3'4'}$ ~5.2 Hz, 1, H3'), 4.40 (m, 3, H4', 5',5''), 3.80 (Δδ 0.015) (2d, J_{gem} ~−11.5 Hz, 1) and 3.56 (Δδ 0.005) (2 d, 1) (6-CH_2OH), 2.10 (Δδ ~0.005) (6 s, 9, OAc's), 1.52 (Δδ 0.01) (2 s, 3, 6-CH_3), D_2O exchanged peaks at δ 5.6 (NH) and 2.9 (OH); ^{13}C NMR (100.6 MHz, $CDCl_3$) δ 170.45, 169.55, 169.49 + 169.41 (CH_3CO's), 146.33 (C2), 134.56 + 134.52 (C4), 131.85 (C8), 122.97 + 122.85 (C5), 86.21 + 86.18 (C1'), 79.94 + 79.85 (C4'), 73.61 + 73.56 (C2'), 70.81 + 70.70 (C3'), 70.44 (6-CH_2OH), 63.31 (C5'), 59.01 + 58.94 (C6), 26.32 (6-CH_3), 20.71, 20.46, 20.43 (CH_3CO's); EI mass spectrum (250 °C), m/z 393.1410 (14.4%, M − CH_2OH, calcd for $C_{17}H_{21}N_4O_7$ 393.1410); FAB MS m/z 425 (M + H), 447 (M + Na), 463 (M + K).

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Registry No. **1b**, 15981-63-2; **2b** (isomer 1), 88452-90-8; **2b** (isomer 2), 88475-71-2; **3b**, 88313-94-4; **5b**, 52921-35-4; **6b** (isomer 1), 96482-72-3; **6b** (isomer 2), 96482-73-4; MeOH, 67-56-1; inosine, 58-63-9; 2',3',5'-tri-*O*-acetylinosine, 3181-38-2; 9-(2',3',5'-tri-*O*-acetyl-β-D-ribofuranosyl)-6-chloropurine, 5987-73-5.