of water and acidified with acetic acid to precipitate the product as colorless solid. An analytical sample was obtained by crystallization from DMF–CH_3CN (2.15 g, 76%): mp 215 °C; ^1H NMR ($(CD_3)_2SO$) δ 10.66 (s, 1, NH, exchangeable with D_2O), 9.1 (s, 1, NH, exchangeable with D_2O), 7.97 (s, 1, 2-H), 7.26 (m, 5, phenyl protons), 5.2 (s, 1, 7-H), 4.76 (s, 2, CH_2), 3.0 (s, 3, CH_3); ^{13}C NMR ($(CD_3)_2SO$) δ 36.1 (CH_3), 51.5 (CH_2), 80.7 (C7), 127.4, 128.8 (aromatic), 152.7 (C4 or C9a), 157.9 (C9a or C4), 161.5 (C6 or C8), 163.25 (C8 or C6), 163.6 (C2); IR (KBr) 3225 m, 3000 m, 1700 vs, 1580 vs, 1370 s, 1320 m, 1230 m, 1170 m, 1100 m, 1025 m, 880 s, 800 s, 750 w, 700 w, 490 cm^{-1} m; mass spectrum (70 eV), *m/e* (relative intensity) 282 (M^+, 100); UV (EtOH) λ_{max} (ϵ) 330 nm (4800), 275 (31 355), 262 (31 355). Anal. Calcd for $C_{14}H_{14}N_6O$: C, 59.56; H, 5.00; N, 29.77. Found: C, 59.76; H, 5.02; N, 29.41.

***N*-Carbamoyl-*N'*-[6-(benzylmethylamino)-3,4-dihydro-4-oxopyrimid-2-yl]formamidine (18)** was prepared from **17** in 91% yield following the procedure described for **10**: mp 239–241 °C dec; ^1H NMR ($(CD_3)_2SO$) δ 9.08 (s, 1, formamidine CH), 7.23 (br, 5, aromatic CH's), 5.0 (s, 1, 5-H), 4.7 (m, 2, CH_2), 2.96 (s, 3, CH_3), 8.0, 8.7, 10.6 (br, NH's); mass spectrum (70 eV), *m/e* (relative intensity) 301 ($M^+ + 1$, 7.5), 300 (M^+, 41), 299 ($M^+ - 1$, 4), 283 ($M^+ - 17$, 10), 282 ($M^+ - 18$, 48), 267 ($M^+ - 33$, 47), 257 ($M^+ - 43$, 9), 254 ($M^+ - 46$, 10), 253 ($M^+ - 47$, 38), 240 ($M^+ - 60$, 17), 215 ($M^+ - 85$, 12), 191 ($M^+ - 109$, 22), 162 ($M^+ - 148$, 30), 120 ($M^+ - 180$, 55). Anal. Calcd for $C_{14}H_{16}N_6O_2$: C, 55.99; H, 5.37; N, 27.99. Found: C, 55.67; H, 5.51; N, 27.65.

Acknowledgment. This research was supported by Research Grant No. CHE 81-21796 from the National Science Foundation and in part by an unrestricted grant from Eli Lilly and Company. We are grateful to the students in a course in X-ray crystallography at the University of Illinois who participated in the single-crystal structure analysis of the pivotal compound **5a** as a class exercise and to Robert C. Hall who was their teaching assistant. High-resolution mass spectral data were obtained in part under a grant from the National Institute of General Medical Sciences (GM-27029). NMR data were obtained in part with support from the University of Illinois NSF Regional Instrumentation Facility, Grant NSF CHE 79-16100.

Registry No. **1**, 108-53-2; **2a**, 51688-22-3; **2b**, 96427-28-0; *syn*-**3a**, 96575-94-9; *anti*-**3a**, 96575-69-8; **3b**, 96575-75-6; **3c**, 96575-74-5; **3d**, 96575-79-0; **4a**, 96575-70-1; **4a**·HCl, 96575-71-2; **4b**, 96575-76-7; **5a**, 96575-73-4; **5b**, 96575-78-9; **6a**, 76299-85-9; **6a**·Na, 96575-80-3; *syn*-**7**, 96575-95-0; *anti*-**7**, 96575-81-4; **8**, 96575-82-5; **8**·HCl, 96575-83-6; **9**, 96575-93-8; **10**, 96575-84-7; **11**, 3977-29-5; **12**, 96575-85-8; **12**·Na, 96575-86-9; **13**, 96575-87-0; **14**, 96575-88-1; **15**, 1194-21-4; **16**, 37409-94-2; **17**, 96575-89-2; **17**·Na, 96575-90-5; **18**, 96575-91-6; $CF_3SO_2OSi(CH_3)_3$, 27607-77-8; H_2NCN, 420-04-2; $C_6H_5CH_2NHCH_3$, 103-67-3.

Nucleoside Annelating Reagents: *N*-(*tert*-Butoxycarbonyl)-2-bromoacetamide and 2-Chloroketene Diethyl Acetal

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The title reagents undergo cyclocondensation reactions with the amidine-like moiety of tri-*O*-acetyladenosine and cytidine (and related compounds) to form a new five-membered ring (etheno bridging), substituted with a (*tert*-butoxycarbonyl)amino or an ethoxy function. Some of the products exhibit useful fluorescence properties. Removal of the *tert*-butoxycarbonyl group with standard conditions gives the corresponding, somewhat unstable amino compounds which can be conveniently characterized as the corresponding *N*-acetyl derivatives. The reagents introducing additional substitution on the etheno bridge, with enhanced fluorescence, also suggest the possibility of cross-linking functionalization.

The discovery that the reaction of adenosine and its derivatives with chloroacetaldehyde in aqueous solution leads to fluorescent products that can be used for the spectroscopic investigation of coenzyme–enzyme interactions and nucleic acid–protein interactions has contributed immensely to our present knowledge of how these biological macromolecules interact.[1] Our continuing program of synthesizing modified nucleosides that provide defined dimensional alterations of conventional enzyme substrates and/or highly fluorescent analogues has led us to investigate the reactions of 2-bromoacetamide carbamates (**1a–c**) and 2-chloroketene diethyl acetal (**2**) with representative adenosine and cytidine derivatives.

1a R = *t*-Bu
b R = Me
c R = CH_2Ph

2

(1) (a) Leonard, N. J. *CRC Crit. Rev. Biochem.* **1984**, *15*, 125 and references therein. [Note: interchange pages 157 and 158.] (b) Leonard, N. J. *Acc. Chem. Res.* **1982**, *15*, 128 and references therein.

Reactions with 2-Bromoacetamide Carbamates. *N*-(*tert*-Butoxycarbonyl)-2-bromoacetamide (**1a**) is a stable colorless crystalline solid that is readily prepared in multigram quantities by the action of oxalyl chloride on 2-bromoacetamide and subsequent quenching of the putative *N*-carbonyl chloride or acylisocyanate intermediate with *tert*-butyl alcohol.[2,3] Benzyl alcohol or methanol may also be used as the quenching agent, thus making it possible to prepare reagents in which the latent amine is masked by carboxylate groups which can be removed under acidic (*t*-Bu, **1a**), basic (Me, **1b**) or reductive ($PhCH_2$, **1c**) conditions.[4] In practice, we discovered that all three reagents **1a–c** will undergo a cyclocondensation reaction

(2) This procedure is a modification of that described in ref 3a for the preparation of $ClCH_2CONHCO_2CH_3$.

(3) (a) Bochis, R. J.; Dybas, R. A.; Eskola, P.; Kulsa, P.; Linn, B. O.; Lusi, A.; Meitzner, E. P.; Milkowski, J.; Mrozik, H.; Olen, L. E.; Peterson, L. H.; Tolman, R. L.; Wagner, A. F.; Waksmunski, F. S.; Egerton, J. R.; Ostlind, D. A. *J. Med. Chem.* **1978**, *21*, 235. See also ref 3b and 3c: (b) Bochis, R. J.; Olen, L. E.; Fisher, M. H.; Reamer, R. A.; Wilks, G.; Taylor, J. E.; Olson, G. *Ibid.* **1981**, *24*, 1483. (c) Bochis, R. J.; et al. *Ibid.* **1981**, *24*, 1518. See also: (d) Speziale, A. J.; Smith, L. B.; Fedder, J. E. *J. Org. Chem.* **1965**, *30*, 4306.

(4) Greene, T. W. "Protective Groups in Organic Synthesis"; Wiley-Interscience: New York, 1981; p 218.

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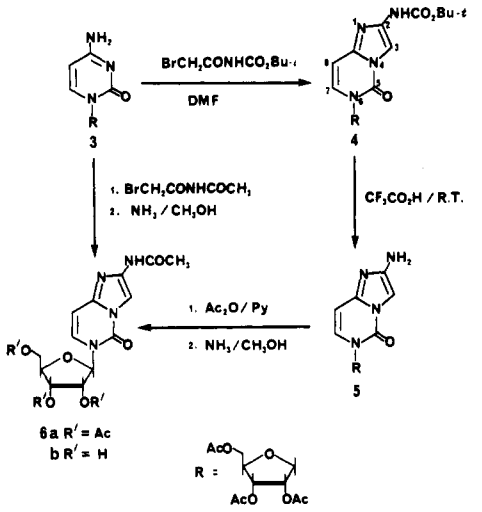
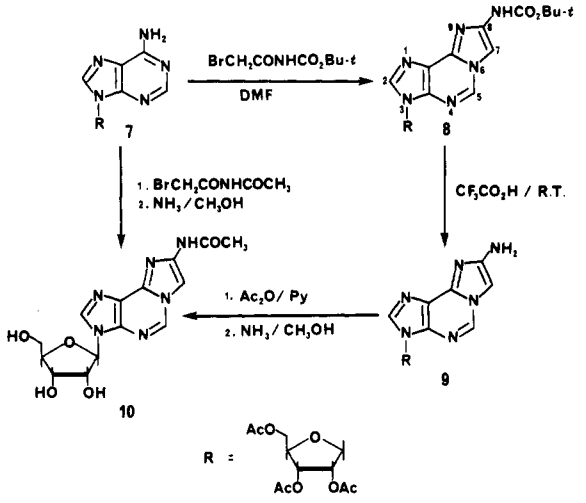
Scheme I



Scheme II



with nucleoside substrates. The acid lability and convenience of removal of the *tert*-butoxycarbonyl (BOC) group made **1a** the reagent of choice for our particular application.

We found that 2′,3′,5′-tri-*O*-acetylcytidine (**3**, Scheme I) required heating at 80 °C in dimethylformamide (DMF) or dimethylacetamide (DMA) solution to undergo reaction with reagent **1a** at a significant rate, whereas 2′,3′,5′-tri-*O*-acetyladenosine (**7**, Scheme II) reacted best at 40–45 °C. In both cases, the times and temperatures cited are those estimated (TLC) to give optimum product formation. The use of a large excess (5–10 mol equiv) of **1a**, elevated temperature, or prolonged reaction time did not significantly increase the yield and, in the case of the latter two variables, resulted in lower yields due to extensive side product formation. The highest yields were obtained when freshly dried solvent was used.

The carbamates **4** and **8** were isolated as TLC-homogeneous syrups after an aqueous workup followed by column chromatography in yields of 38 and 52%, respectively, and were characterized by ^{1}H NMR and high- and low-resolution mass spectrometry. That a cyclocondensation reaction to give a new five-membered ring had occurred in each case was suggested by the absence of the broad resonance due to the exocyclic amino function, the existence of an additional one-proton singlet at δ 7.45 (**4**) and 7.90 (**8**), and the presence of a broad exchangeable resonance (one proton) at δ 9.92 (**4**) and 10.08 (**8**) in the ^{1}H NMR spectra measured in $(CD_3)_2SO$ solution. The new singlet resonances at 7.45 and 7.90 ppm did not diminish in intensity upon brief treatment with D_2O. In $CDCl_3$ solution these resonances appeared unusually broad (peak width at half-height ca. 6 Hz) in both cases, but the signals did not diminish appreciably in intensity when C_2H_5OD was added.

The proposed regiochemistry of the cyclocondensation products is supported by analogy with results obtained elsewhere.[5a] In addition, the presumed mechanism of the reaction in which the carbonyl adjacent to the halomethylene group condenses with the exocyclic amine and the halomethylene alkylates the endocyclic nitrogen also suggests the regiochemistry as shown.[5b]

Treatment of **4** or **8** with anhydrous trifluoroacetic acid (TFA) at room temperature cleanly and rapidly removed the BOC group. Direct evaporation of the reaction mixture gave the amine trifluoroacetate salts which underwent decomposition at room temperature. However, if an aqueous workup employing an extraction with 1 M aqueous K_2CO_3 solution was used instead, the free amines **5** and **9** were isolated in moderate yields.

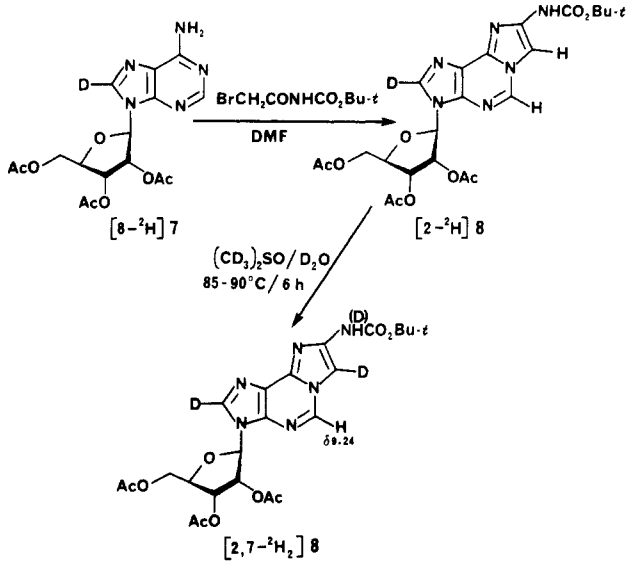
Compound **5** was isolated as a crystalline solid which could be stored at room temperature for several weeks without appreciable decomposition; however, compound **9** was considerably less stable and was best stored at −20 °C in the dark. The observation that the resonance assigned to 3-H in compound **5** and 7-H in compound **9** underwent rapid deuterium exchange when treated with D_2O in $(CD_3)_2SO$ solution was a characteristic feature of the ^{1}H NMR spectra of these substances. For complete characterization, the amines were converted by N-acetylation followed by selective de-*O*-acetylation into the stable, colorless, crystalline mono-*N*-acetyl derivatives **6b** and **10**. They were found to be identical in chromatographic and physical properties with the compounds prepared by treatment of **3** and **7**, respectively, with (1) $BrCH_2CONHCOCH_3$ and (2) NH_3/MeOH (Schemes I and II). We should state at this juncture that the *N*-acetyl group on molecules **6b** and **10** was resistant to removal under the usual acidic or basic (NH_3/MeOH) conditions, which meant that we were unable to prepare the free amines **5** and **9** by this alternate route.

It should be noted that the BOC group does not function solely as a protecting group. The electronic effect of the group is such that the carbonyl adjacent to the $BrCH_2$ moiety becomes less amide-like, thus activating the carbonyl toward condensation with a nucleoside exocyclic primary amino function. The observation that 2-bromoacetamide itself underwent no annelation reactions with tri-*O*-acetylcytidine, adenosine, or guanosine but reacted instead by alkylation at a ring N serves to emphasize this point. In the case of tri-*O*-acetylguanosine, the site of N7-alkylation was confirmed by the observation that the proton at the 8-position in the alkylated product underwent rapid deuterium exchange when treated with D_2O in $(CD_3)_2SO$ solution. A similar phenomenon has been observed by Eistetter and Pfleiderer.[6]

It was possible to make unequivocal assignments in the ^{1}H NMR spectra of the cytidine-derived compounds (Scheme I) (see Table II, Experimental Section) since the chemical shift data combined with signal multiplicity provided sufficient information. The generalities indicated

(5) (a) Barrio, J. R.; Sattsangi, P. D.; Gruber, B. A.; Dammann, L. G.; Leonard, N. J. *J. Am. Chem. Soc.* **1976**, *98*, 7408. (b) Ref 1a, p 128.
(6) Eisetter, K.; Pfleiderer, W. *Chem. Ber.* **1973**, *106*, 1389. See also ref 7.
(7) Brookes, P.; Lawley, P. D. *J. Chem. Soc.* **1961**, 3923.

Scheme III



by the ^1H NMR spectra of **3**, **4**, **5**, and **6a** are as follows: annelation of the five-membered ring onto the cytidine nucleus causes a marked downfield chemical shift for the original 5-H, a concomitant but small upfield shift for the original 6-H, and accordingly a decrease in the chemical shift difference between the pyrimidine ring protons.

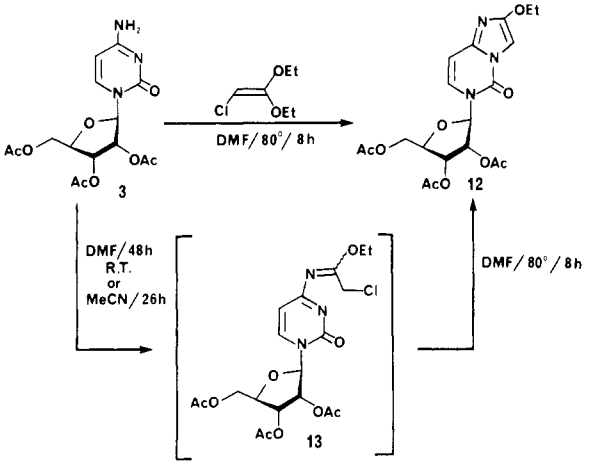
Assignments in the ^1H NMR spectra of the adenosine-derived compounds (Scheme II) were made unambiguously by means of deuterium labeling experiments (see Table I, Experimental Section). For example, the chemical shift for the proton on the ribosyl-substituted imidazole ring was identified by initial 8-deuterium labeling of 2′,3′,5′-tri-*O*-acetyladenosine ([8-^2H]**7**) and subsequent reaction with *N*-(*tert*-butoxycarbonyl)-2-bromoacetamide (**1a**) to give [2-^2H]**8** (Scheme III). The proton on the newly formed imidazole ring was then differentiated from the proton on the pyrimidine ring by deuterium exchange of the former at raised temperature in $(CD_3)_2SO/D_2O$ to give [2,7-$^2H_2$]**8**. A useful generality indicated by the ^1H NMR spectra of **8**, **9**, and **10**, as measured in $(CD_3)_2SO$ solution, is that annelation of the five-membered ring onto the adenosine nucleus causes a marked downfield shift for the pyrimidine ring proton.

Reactions with 2-Chloroketene Diethyl Acetal (2). The reaction of simple heterocyclic amines with 2-chloroketene diethyl acetal (**2**) has been examined by Kato, Yamamoto, and Takeda.[8] For example, the heating of 2-aminopyridine with **2** in the absence of solvent at 100 °C led to the formation of 2-ethoxyimidazo[1,2-*a*]pyridine (8% yield) and 2-(2-pyridylamino)imidazo[1,2-*a*]pyridine (17% yield) as the major products.[8] We found that compound **2** reacted in DMF solution (preferred solvent) with the tri-*O*-acetyl derivatives of adenosine and cytidine to yield the products **11** and **12**, respectively. Four equivalents of **2** were used to encourage complete conversion of starting material; with 1.0 or 2.0 equiv of **2**, significant amounts of starting materials remained even after prolonged reaction time and/or raised temperature.

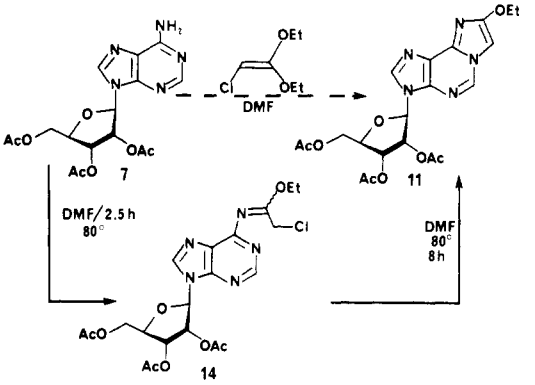
Tri-*O*-acetylcytidine (**3**) reacted with compound **2** at room temperature in either anhydrous DMF or acetonitrile to give mainly (TLC, ^1H NMR) N^4-(1-ethoxy-2-chloroethylidene)-2′,3′,5′-tri-*O*-acetylcytidine (**13**) along with a small amount of the cyclized compound (**12**). It is not necessary to isolate the intermediate **13** since **3** can be

(8) Kato, T.; Yamamoto, Y.; Takeda, S. *Yakugaku Yasshi* **1974**, *94*(5), 627; *Chem. Abstr.* **1974**, *81*(7), 37514u.

Scheme IV



Scheme V



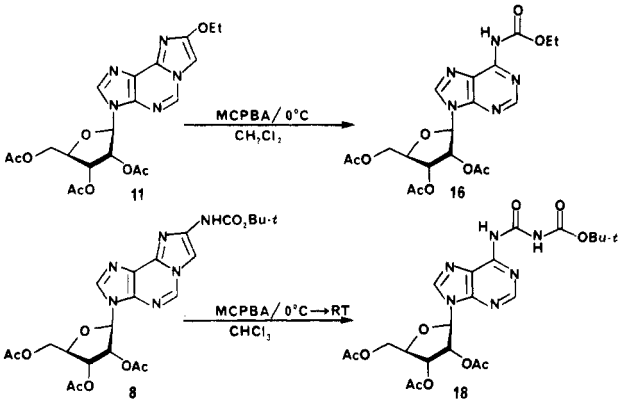
cleanly converted to **12** in one step (Scheme IV) by heating at 80 °C in DMF solution. The product, 2-ethoxy-5,6-dihydro-5-oxo-6-(2′,3′,5′-tri-*O*-acetyl-β-D-ribofuranosyl)-imidazo[1,2-*c*]pyrimidine (**12**), was isolated as a viscous oil in 85% yield.

In the case of tri-*O*-acetyladenosine (**7**) a two-step procedure was found to give best results; isolation of the intermediate N^6-(1-ethoxy-2-chloroethylidene)-2′,3′,5′-tri-*O*-acetyladenosine (**14**) followed by dissolution in anhydrous DMF and heating at 80 °C for 8 h induced cyclization (Scheme V). In this way, the highly fluorescent tricyclic nucleoside **11** was obtained as a noncrystalline viscous oil in 39% yield. Tri-*O*-acetylguanosine reacted with **2** to give the N^2-(1-ethoxy-2-chloroethylidene)-2′,3′,5′-tri-*O*-acetylguanosine as a pale-violet foam in 74% yield, but upon attempted cyclization in anhydrous DMF at 80 °C for 24 h, only decomposition to tri-*O*-acetylguanosine was observed.

It is interesting to compare the reactivities of compounds **13** and **14**. The former shows a markedly increased propensity for cyclization compared with compound **14**. For example, when **13** was subjected to the conditions (80 °C/DMF/2.7 h) used to prepare **14**, ca. 60% of the material (TLC) was converted to the cyclized compound **11**.

The reaction between 2-chloroketene diethyl acetal (**2**) and 1-*n*-propylcytosine produced crystalline 2-ethoxy-5,6-dihydro-5-oxo-6-*n*-propylimidazo[1,2-*a*]pyrimidine (**15**) which was characterized by elemental analysis and spectroscopy. Compound **15** was very reactive toward electrophiles. Numerous compounds resulted from treatment of **15** with *N*-bromosuccinimide (NBS) or *N*-chlorosuccinimide (NCS) in $CHCl_3$ or CCl_4 solution. Treatment of **15** at 0 °C with *m*-chloroperbenzoic acid (MCPBA) in dichloromethane solution resulted in a rapid oxidative

Scheme VI



cleavage of the five-membered ring to give 4-*N*-(ethoxycarbonyl)-1-*n*-propylcytosine (**17**) as the major product along with several minor components. This product decided the direction of the ring closure as leading to **15**. The reaction worked best with 3 mol equiv of oxidizing agent for the complete conversion of starting material. Apparently, in its reaction with MCPBA, **15** behaves not as a fully conjugated heterocyclic system but rather as an electron-rich olefin appended to a cytidine nucleus since the reaction probably proceeds by initial epoxidation of the ethyl enol ether functionality. A similar reaction occured with **11** giving the ethyl carbamate derivative **16** as the major product (Scheme VI). Additionally, compound **8** underwent oxidation with MCPBA in chloroform solution to give the urea **18** as the major product in 30% yield (not optimized) (Scheme VI).

The preparation of compound **10** in pure crystalline form enabled us to determine the fluorescence characteristics of a $1,N^6$-ethenoadenosine substituted with an acetylamino function at the 8-position of the etheno bridge. The fluorescence intensity of this compound, like that of ϵAdo, is extremely high, and the substitution leads to a higher quantum yield (Φ = 0.81) and longer emission wavelength (λ 421 nm in pH 7 phosphate buffer, excitation at 245 nm) than for ϵAdo (unsubstituted on the etheno bridge). At pH 2 or 11 the fluorescence of **10** is ca. 15% of that at pH 7.

We have described in this paper how reagent **1a** reacts with nucleoside substrates to generate, regiospecifically, over two steps and under mild conditions the sensitive primary enamine functionality on a complex natural product. We believe that this methodology is a valuable extension of the Tschitshibabin reaction[9] and as such should find general applicability in heterocyclic synthesis. Reagent **2** also reacts regiospecifically with certain nucleoside substrates to generate the reactive ethyl enol ether functionality. This reaction should also find general applicability in the transformation of heterocyclic systems.

Experimental Section

Melting points were determined on a Thomas-Hoover capillary melting point apparatus and are uncorrected. Microanalyses were carried out by Josef Nemeth and his staff at the University of Illinois. ^{1}H NMR spectra were measured at 360 MHz on a Nicolet NTC-360 spectrometer or at 90 MHz with a Varian EM390 spectrometer. Tetramethylsilane was used as internal standard in all NMR spectra, and the following abbreviations are used: s singlet, d doublet, t triplet, m multiplet, br broad, and ex exchangeable with D_2O.

Mass spectra (MS) were obtained by three different techniques: fast atom bombardment (FAB) with a VG ZAB-1 HF mass spectrometer, field desorption (FD) with a Varian MAT-731 mass spectrometer, or electron impact (EI) with a Varian MAT CH-5 mass spectrometer. Mass spectral data are quoted as *m/e* (relative intensity). Infrared (IR) spectra were recorded on a Perkin-Elmer 337 grating infrared spectrophotometer; br broad and sh shoulder are the only abbreviations used. Ultraviolet (UV) absorption spectra were measured on a Beckman Acta MVI spectrophotometer; sh shoulder, w weak, m medium, and s strong are the abbreviations used to describe qualitative spectra. Technical fluorescence excitation and emission spectra were obtained by photon counting on a Spex Fluorolog 111C spectrofluorimeter coupled with a Datamate microprocessor.

Merck silica gel 60F-254 precoated glass-backed plates were used for thin-layer chromatography (TLC). The following solvent systems were used: A, chloroform–methanol (19:1, v/v); B, chloroform–methanol (9:1, v/v); C, chloroform–methanol (4:1, v/v). After development, substances were detected by UV light. Brinkmann silica gel (0.05–0.2 mm) was used for column chromatography. PLC (preparative-layer chromatography) was carried out with Merck silica gel 60 F_{254} precoated plates (20 × 20 cm, 2 mm thickness).

DMF and DMA were purified by stirring with powdered CaH_2 for 16 h followed by distillation under reduced pressure. Pyridine was dried by heating under reflux with CaH_2 followed by distillation at atmospheric pressure. All other solvents and reagents were reagent grade unless otherwise specified.

***N*-(*tert*-Butoxycarbonyl)-2-bromoacetamide (1a).** 2-Bromoacetamide was prepared in 92% yield by bubbling ammonia gas through a mechanically stirred benzene solution of 2-bromoacetylbromide (Eastman Kodak Co.; 1 g/10 mL of C_6H_6) until the reaction mixture was alkaline to wet litmus, followed by evaporation of the reaction mixture and extraction of the amide into boiling benzene, mp 89–91 °C (lit.[10] mp 91 °C). A stirred suspension of 2-bromoacetamide (22.65 g, 164 mmol) in 1,2-dichloroethane (120 ml) in a three-necked flask, fitted with rubber septa and a reflux condenser attached to a N_2 line, was cooled (ice–water bath) while oxalyl chloride (Gold Label, 25.0 g, 197 mmol) was added by syringe. The mixture was stirred for 10 min and then allowed to warm to room temperature. The rubber septa were replaced by glass stoppers, a paraffin oil bubbler was attached to the top of the condenser (gas evolution), and the mixture was brought to reflux. After 2.5 h at reflux, the reaction mixture was cooled (ice–water bath) while a mixture of *tert*-butyl alcohol and 1,2-dichloroethane (1:1, v/v, 65 mL) was added in one portion. After 15 min the cooling bath was removed and the reaction mixture was poured into ice-cold saturatred sodium bicarbonate solution (500 mL) with the aid of 300 mL of 1,2-dichloroethane for rinsing. The organic phase was extracted rapidly, fresh sodium bicarbonate solution was added, and the process was repeated. The organic layer was washed with water (400 mL), dried ($MgSO_4$), and evaporated in vacuo to give an orange oil (37 g) which crystallized slowly. Recrystallization from cyclohexane (200 mL) with decolorizing carbon treatment and hot filtration gave 24.3 g of product, mp 106–107 °C. Concentration of the mother liquors and recrystallization from cyclohexane gave a further 3.6 g: mp 102–105 °C (total yield 71%); ^{1}H NMR ($CDCl_3$) δ 1.50 (s, 9, *t*-Bu), 4.27 (s, 2, CH_2Br); IR (KBr) 3250, 3195, 2990 (br), 1765 (sh), 1750, 1530, 1400, 1370, 1250, 1145, 1040, 1010, 990, 850, 785 cm^{-1}; MS (EI, 70 eV), 181 (M^+ – *t*-Bu, 5.6), 137 (0.5), 123 (3.6), 121 (3.5), 102 (1.8), 95 (4.3), 93 (4.1), 59 (40), 57 (100), 43 (15), 42 (11.5), 41 (49.9). Anal. Calcd for $C_7H_{12}BrNO_3$: C, 35.32; H, 5.08; N, 5.88; Br, 33.56. Found: C, 35.26; H, 5.14; N, 5.87; Br, 33.52.

Also prepared in a similar manner was ***N*-(methoxycarbonyl)-2-bromoacetamide (1b)**: mp 152–154 °C (1,2-dichloroethane); yield 81%; ^{1}H NMR [$(CD_3)_2SO$, 90 MHz] δ 3.70 (s, 3, OCH_3), 4.20 (s, 2, CH_2Br); IR (KBr) 3250, 3175, 3000 (br), 1765, 1535, 1400, 1235, 1200, 1175, 1050, 900, 785, 620 cm^{-1}. Anal. Calcd for $C_4H_6BrNO_3$: C, 24.47; H, 3.08; N, 7.15; Br, 40.65. Found: C, 24.63; H, 3.04; N, 7.08; Br, 40.39.

***N*-(Benzyloxycarbonyl)-2-bromoacetamide (1c)**: mp 142–143 °C (benzene); yield 67%; ^{1}H NMR [$(CD_3)_2SO$, 90 MHz] δ 4.19 (s, 2, CH_2Br), 5.14 (s, 2, CH_2), 7.38 (m, 5, Ph); IR (KBr) 3250, 3175, 3000 (br), 1765, 1545, 1400, 1225, 1180, 1055, 1025,

(9) Tschitschibabin, A. E. *Ber. Dtsch. Chem. Ges.* **1925**, *58*, 1704.

(10) Bischoff, C. A. *Ber. Dtsch. Chem. Ges.* **1897**, *30*, 2311.

925, 785, 760, 595 cm^{-1}. Anal. Calcd for $C_{10}H_{10}BrNO_2$: C, 44.13; H, 3.70; N, 5.15; Br, 29.37. Found: C, 44.16; H, 3.72; N, 5.08; Br, 29.54.

2-[*N*-(*tert*-Butoxycarbonyl)amino]-5,6-dihydro-5-oxo-6-(2′,3′,5′-tri-*O*-acetyl-β-D-ribofuranosyl)imidazo[1,2-*c*]pyrimidine (4). A stirred solution of 2′,3′,5′-tri-*O*-acetylcytidine[11] (3.229 g, 8.75 mmol) and *N*-(*tert*-butoxycarbonyl)-2-bromoacetamide (3.124 g, 13.13 mmol) in dry DMF (30 mL) was heated at 70–75 °C (bath temperature) for 7 h under an atmosphere of N_2. During this time, the reaction mixture was monitored by TLC (system C) for the progressive formation of the product which had a considerably higher R_f when compared with the starting material. The dark-red solution was evaporated in vacuo, and the residue was dissolved in $CHCl_3$ (120 mL), washed with 1 M NaOH (3 × 100 mL) and then H_2O (2 × 100 mL), dried ($MgSO_4$), and evaporated to give a thick syrup (1.75 g). The syrup was purified by column chromatography on silica gel (60 g). Gradient elution with methanol in chloroform (0 → 4%, v/v) gave the product as an orange syrup (1.70 g, 38%): R_f (system B) 0.69; ^1H NMR ($CDCl_3$, 90 MHz) δ 1.55 (s, 9, *t*-Bu), 2.05, 2.10, 2.14 (3 s, 9, $COCH_3$), 4.37 (br s, 3, 4′-H, 5′-H), 5.39 (m, 2, 2′-H, 3′-H), 6.29 (m, 1, 1′-H), 6.69 (d, *J* = 8 Hz, 1, 8-H), 7.18 (d, *J* = 8 Hz, 1, 7-H), 7.73 (br s, 1, 3-H), 9.10 (br, 1, NH, ex); high-resolution MS (FAB) obsd 509.1864, $C_{22}H_{29}N_4O_{10}$ 509.1884; low-resolution MS (FAB) 509 (MH^+, 12), 453 (15), 408 (6), 259 (34), 195 (100), 151 (38), 139 (69), 97 (25), 85 (12), 69 (6), 57 (23), 43 (37).

2-Amino-5,6-dihydro-5-oxo-6-(2′,3′,5′-tri-*O*-acetyl-β-D-ribofuranosyl)imidazo[1,2-*c*]pyrimidine (5). The foregoing BOC-protected nucleoside (4) as a syrup (1.70 g, 3.35 mmol) was dissolved in a minimum amount of neat TFA by swirling. The solution was kept at room temperature and protected from moisture for 5–10 min, after which time TLC (system C) indicated that no starting material was present and one major new substance of much lower R_f was detected. The TFA was then removed in vacuo (bath temperature ~35 °C.) The gummy residue was dissolved in $CHCl_3$ (200 mL), washed with 1 M aqueous K_2CO_3 solution (2 × 105 mL) and H_2O (1 × 100 mL), dried ($MgSO_4$), and evaporated in vacuo to give a thick gum that slowly crystallized to give a pale-orange solid (0.80 g, 59%). The substance decomposed on attempted recrystallization from a variety of solvents and also on attempted purification by silica gel chromatography with chloroform–methanol as the eluting solvent; however, the physical and spectroscopic data indicated that it was sufficiently pure to be used in subsequent transformations: mp 135–138 °C dec; begins to darken at 110 °C; ^1H NMR [$(CD_3)_2SO$, 90 MHz] δ 2.11 (s, 6, $COCH_3$), 2.16 (s, 3, $COCH_3$), 4.29 (br s, 3, 5′-H, 4′-H), 5.04 (m, 2, NH_2 ex), 5.43 (m, 2, 2′-H, 3′-H), 6.10 (d, *J* = 5 Hz, 1, 1′-H), 6.40 (d, *J* = 8 Hz, 1, 8-H), 6.62 (s, 1 3-H, ex), 7.40 (d, *J* = 8 Hz, 1 H, 7-H); high-resolution MS (FAB) obsd 409.1363, $C_{17}H_{20}N_4O_8$ 409.1360; low-resolution mass spectrum (FAB) 409 (MH^+, 35), 346 (4), 344 (6), 259 (12), 151 (100), 139 (35), 124 (8); qual UV λ_{max} (MeOH) 308 nm (m) 233 (s); R_f (system C) 0.62. The substance had a characteristic blue appearance when a freshly developed thin-layer chromatogram was observed under short wavelength UV light.

2-(Acetylamino)-5,6-dihydro-5-oxo-6-(2′,3′,5′-tri-*O*-acetyl-β-D-ribofuranosyl)imidazo[1,2-*c*]pyrimidine (6a). Method A. *N*-(Bromoacetyl)acetamide was prepared by acetylation of 2-bromoacetamide with acetic anhydride (1.4 ml/mmol) and concentrated sulfuric acid (3.5 mg/mmol) at room temperature for 36 h: yield 77%; mp 118–120.5 °C. A solution of 2′,3′,5′-tri-*O*-acetylcytidine[11] (0.180 g, 0.487 mmol) and *N*-(bromoacetyl)acetamide (0.135 g, 0.75 mmol) in dry DMA (3 mL) was stirred and heated at 80 °C (bath temperature) under an atmosphere of N_2. After 4 h more *N*-(bromoacetyl)acetamide (0.090 g, 0.5 mmol) was added and the reaction was continued for a further 1 h. The progressive formation of the product (R_f 0.68, system C) could be followed readily by thin-layer chromatographic examination of the reaction mixture. The solvent was evaporated in vacuo and the dark-violet residue was applied to a column of silica gel (15 g). Elution with methanol in chloroform (0 → 4%, v/v) evaporation of the appropriate fractions, and trituration with ether gave the pure product (0.053 g, 38% based on unrecovered starting material) as an amorphous solid. Further elution gave unreacted starting material (0.065 g, 36%): ^1H NMR ($CDCl_3$, 360 MHz) δ 2.087, 2.138, 2.155, 2.189 (4 s, 12, $COCH_3$), 4.40 (br s, 3, 5′-H, 4′-H), 5.39–5.43 (m, 2, 2′-H, 3′-H), 6.30 (d, *J* = 5.3 Hz, 1, 1′-H), 6.52 (d, *J* = 7.9 Hz, 1, 8-H), 7.24 (d, *J* = 7.9 Hz, 1, 7-H), 8.08 (s, 1, 3-H), 9.23 (s, 1, NH).

Further characterization was facilitated by selective de-O-acetylation [methanolic ammonia (half-saturated at 0 °C)/room temperature/6 h] to the **mono-*N*-acetyl derivative (6b)**, which crystallized directly from the reaction medium: mp 235–237 °C dec; yield 63%; high-resolution MS (FAB) obsd 324.1053, $C_{13}H_{16}N_4O_6$ 324.1071; ^1H NMR [$(CD_3)_2SO$, 360 MHz] δ 2.03 (s, 3, $COCH_3$), 3.60 (m, 2, 5′-H), 3.89–4.10 (m, 3, 4′-H, 3′-H, 2′-H), 5.12 (m, 2, OH, ex), 5.43 (d, *J* = 5.5 Hz, 1, OH, ex), 6.02 (d, *J* = 5.1 Hz, 1, 1′-H), 6.62 (d, *J* = 7.8 Hz, 1, 8-H), 7.74 (s, 1, 3-H), 7.76 (d, *J* = 7.8 Hz, 1, 7-H), 10.68 (s, 1, NH, ex); UV λ_{max} (0.08 M NaOH) 286 nm (ε 12 800), 238 (sh) (23 900); λ_{max} (0.08 M HCl) 297 nm (ε 11 500), 225 (20 000); fluorescence λ_{max}^{em} (0.1 M HCl) 357 nm; fluorescence λ_{max}^{ex} 305 nm. At pH 7 or 11 the fluorescence intensity is ~10% of that at pH 2. Anal. Calcd for $C_{13}H_{16}N_4O_6 \cdot CH_3OH$: C, 47.19; H, 5.66; N, 15.72. Found: C, 47.35; H, 5.34; N, 16.06. (Obtained after a 3-fold recrystalliation from methanol and drying at 120 °C for 16 h at 2 torr.)

Method B. 2-Amino-5,6-dihydro-5-oxo-6-(2′,3′,5′-tri-*O*-acetyl-β-D-ribofuranosyl)imidazo[1,2-*c*]pyrimidine (5, 0.346 g, 0.848 mmol) was dissolved in dry pyridine (2 mL) and acetic anhydride (1 mL) was added. After 16 h at room temperature the reaction was quenched by the addition of methanol (1 mL), and the mixture was stirred for 10 min and then evaporated in vacuo. After coevaporation with toluene (4 × 25 mL), the residue was dissolved in $CHCl_3$ (50 mL), washed with saturated sodium bicarbonate solution (50 mL) and water (2 × 50 mL), dried ($MgSO_4$), and evaporated in vacuo to give a yellow gum. The gum was dissolved in methanolic ammonia (half-saturated at 0 °C, 6 mL), left at room temperature for 5 h, then stored in the refrigerator for a further 16 h, which caused the product to crystallize from solution. The crystals were collected and dried in vacuo to give 0.155 g (56%) of compound **6b**, mp 238–240 °C dec. The substance had physical and spectroscopic properties identical with those of the compound prepared by method A.

8-[*N*-(*tert*-Butoxycarbonyl)amino]-3-(2′,3′,5′-tri-*O*-acetyl-β-D-ribofuranosyl)imidazo[2,1-*i*]purine (8). A stirred solution of 2′,3′,5′-tri-*O*-acetyladenosine[12] (2.358 g, 6.0 mmol) and *N*-(*tert*-butoxycarbonyl)-2-bromoacetamide (1a) (1.428 g, 6.0 mmol) in dry DMF (40 mL) was heated at 40–45 °C (bath temperature) for 90 h under an atmosphere of N_2. The dark-red solution was evaporated in vacuo, the residue was dissolved in chloroform (250 mL), extracted with 1 M potassium hydroxide solution 2 × 150 mL) and then with water (150 mL), dried ($MgSO_4$), and evaporated to give a yellow syrup, which was purified by column chromatography on silica gel (120 g). Gradient elution with methanol in chloroform (0 → 1.5%, v/v) gave the highly fluorescent product as a syrup homogeneous by TLC (1.66 g, 52%): R_f (system B) 0.45; ^1H NMR ($CDCl_3$, 90 MHz) δ 1.52 (s, 9, *t*-Bu), 2.09, 2.11, 2.15 (3 s, 9, $COCH_3$), 4.40–4.51 (br s, 3, 5′-H, 4′-H), 5.72 (m, 1, 3′-H), 6.04 (m, 1, 2′-H), 6.27 (d, *J* = 5 Hz, 1,1′-H), 7.61 (br s, 1, NH, exchanged with C_2H_5OD), 7.80 (br s, 1, 7-H), 8.18 (s, 1, 2-H), 8.68 (s, 1, 5-H); high-resolution MS (FAB) obsd 533.2011, $C_{23}H_{29}N_6O_9$, 533.1996; low-resolution mass spectrum (EI, 10 eV) 532 (M^+, 0.4), 432 (2), 334 (3), 259 (10), 174 (0.6), 139 (20), 59 (100), 41 (4); qual UV λ_{max} (MeOH) 315 (sh w), 275 (sh w), 249 (sh s), 242 (s).

8-Amino-3-(2′,3′,5′-tri-*O*-acetyl-β-D-ribofuranosyl)-imidazo[2,1-*i*]purine (9). The BOC group was removed from compound 8 (1.30 g, 2.44 mmol) as described for compound 4. The TFA was then removed in vacuo to give a gummy residue that was dissolved in chloroform (200 mL) and extracted with 1 M aqueous K_2CO_3 solution (2 × 105 mL) followed by water (1 × 100 mL). The combined aqueous extracts were back-extracted with chloroform (150 mL), the organic extracts were combined, dried ($MgSO_4$), and evaporated in vacuo to give a dark orange oil 0.638 g (60%). A portion of the oil (80 mg) was purified further

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by PLC. Elution with $CHCl_3$–MeOH (85:5, v/v) and extraction of the main fluorescent band with $CHCl_3$–MeOH (98:2, v/v) gave the substance as an oil homogeneous by TLC (38 mg); R_f (system B) 0.17; ^1H NMR [$(CD_3)_2SO$, 90 MHz] δ 2.05 (s, 6, $COCH_3$), 2.12 (s, 3, $COCH_3$), 4.21–4.53 (m, 3, 5′-H, 4′-H), 5.24 (br s, 2, NH_2, ex), 5.64 (m, 1, 3′-H), 6.05 (m, 1, 2′-H), 6.30 (d, J = 5 Hz, 1, 1′-H), 7.05 (s, 1, 7-H, ex), 8.43 (s, 1, 2-H), 9.02 (s, 1, 5-H); high-resolution MS (FAB) obsd 433.1475, $C_{18}H_{21}N_6O_7$, 433.1472; low-resolution MS (FAB) 433 (MH^+, 29), 394 (3), 309 (7), 279 (17), 259 (10), 205 (17), 175 (36), 155 (46), 152 (36), 149 (36), 135 (54), 119 (100), 103 (56); qual UV λ_{max} (MeOH) 326 (sh w), 283 (sh m), 245 (s).

8-(*N*-Acetylamino)-3-β-D-ribofuranosylimidazo[2,1-*i*]purine (10). Method A. A solution of 2′,3′,5′-tri-*O*-acetyladenosine (1.572 g, 4.0 mmol) and *N*-(bromoacetyl)acetamide (1.44 g, 8.0 mmol) in dry DMA (30 ml) was stirred at room temperature under an atmosphere of N_2 for 72 h. TLC (system B) revealed the progressive formation of the product, which had a near identical R_f to that of the starting material but which had a characteristic blue color when a thin-layer chromatogram was observed under short wavelength UV light. After the reaction appeared to progress no longer, the solvent was evaporated in vacuo (2 torr, 50–60 °C bath temperature), and the residue was dissolved in $CHCl_3$ (80 mL), washed several times with saturated sodium bicarbonate solution, dried ($MgSO_4$), and evaporated in vacuo to yield a syrup which was purified further by chromatography on silica gel (60 g, methanol in chloroform 0 → 6% v/v as eluant) to give a syrup (1.216 g); ^1H NMR [$(CD_3)_2SO$] revealed that the material was a ca. 1:1 mixture of unreacted 2′,3′,5′-tri-*O*-acetyladenosine and product. Moreover, the mixture proved difficult to separate chromatographically. However, the problem was resolved easily by the simple expedient of selective de-*O*-acetylation in methanolic ammonia (half-saturated at 0 °C, 40 mL) from which compound **10** crystallized directly from solution. After 20 h, the crystals were collected, washed with cold methanol (2 × 5 mL), and dried in vacuo to give 0.650 g (47%) of fine colorless needles. The analytical sample was obtained after recrystallization four times from water and drying in vacuo (2 torr, P_2O_5, 138 °C, 18 h): mp 257–259 °C; ^1H NMR ($(CD_3)_2SO$, 360 MHz) 2.09 (s, 3, $COCH_3$), 3.58 (m, 1, 5a′-H), 3.68 (m, 1, 5b′-H), 3.98 (m, 1, 4′-H), 4.18 (m, 1, 3′-H), 4.58 (m, 1, 2′-H), 5.10 (t, J = 5.3 Hz, 1, OH, ex), 5.26 (d, J = 4.9 Hz, 1, OH, ex), 5.55 (d, J = 5.9 Hz, 1, OH, ex), 6.01 (d, J = 5.4 Hz, 1, 1′-H), 8.17 (s, 1, 7-H), 8.55 (s, 1, 2-H), 9.25 (s, 1, 5-H), 10.87 (s, 1, NH, ex); high-resolution MS (FAB) obsd 349.1253, $C_{14}H_{17}N_6O_5$ 349.1260; UV λ_{max} (pH 7 phosphate buffer) 310 nm (ϵ 2300), 275 (7400), 245 (46 300); λ_{max} (0.1 M HCl) 282 nm (ϵ 9900), 247 (sh) (31 100), 242 (33 700); λ_{max} (0.1 M NaOH) 291 nm (ϵ 5900), 245 (33 700); fluorescence λ_{max}^{em} 421 nm, λ_{max}^{ex} 245 nm (pH 7 phosphate buffer), Φ (quinine sulfate 0.70) 0.81 (pH 7) (at pH 2 or 11 the fluorescence intensity is ~15% of that at pH 7). Anal. Calcd for $C_{14}H_{16}N_6O_5$: C, 48.28; H, 4.63; N, 24.13. Found: C, 48.05; H, 4.44; N, 23.88.

Method B. 8-Amino-3-(2′,3′,5′-tri-*O*-acetyl-3-β-D-ribofuranosyl)imidazo[2,1-*i*]purine (**9**) (0.70 g, 0.172 mmol) was dissolved in dry pyridine (2 mL) and acetic anhydride (1.5 mL) was added. After 2 h at room temperature (during which time the reaction mixture formed a gel) methanol (3 mL) was added, and the mixture was stirred for 10 min and then evaporated in vacuo. The residue was dissolved in chloroform (30 mL), washed with saturated sodium bicarbonate solution (2 × 30 mL) and water (2 × 30 mL), dried ($MgSO_4$), and evaporated in vacuo to give an orange oil that was dissolved in methanolic ammonia (half-saturated at 0 °C, 5 mL). After 5 h, examination of the reaction mixture by TLC showed that de-O-acetylation was complete. The solvent was evaporated, and the residue was triturated with ether (5 mL) and then dissolved in water (5 mL). After treatment with decolorizing carbon and hot filtration, the filtrate was evaporated, coevaporated with ethanol (4 × 5 mL), and finally triturated with ether (5 mL). The pale-brown solid (0.045 g, 75%) appeared as one spot on thin-layer chromatographic analysis (R_f 0.17, system C) identical with compound **10** prepared as in Method A.

Reaction of 2′,3′,5′-Tri-*O*-acetylguanosine with (A) *N*-(Bromoacetyl)acetamide and (B) 2-Bromoacetamide. A. A solution of 2′,3′,5′-tri-*O*-acetylguanosine[12] (1.228 g, 3.0 mmol) and *N*-(bromoacetyl)acetamide (1.080 g, 6.0 mmol) in anhydrous DMA (25 mL) was kept for 3 days at room temperature. TLC analysis of the reaction mixture during this time revealed a progressive disappearance of the substrate to give two substances, both of lower R_f than the starting material, but of similar R_f to each other. The solvent was evaporated in vacuo (2 torr/50 °C), and the residue was dissolved in a minimum amount of $CHCl_3$ and applied to a column of silica gel (80 g). Gradient elution with methanol in chloroform (0 → 12%, v/v) yielded two fractions, the first (0.605 g, colorless solid after trituration with ether) was enriched in the higher R_f substance, the second (0.742 g, sticky solid) was enriched in the lower R_f substance. The ^1H NMR spectra of both fractions were similar: ^1H NMR [$(CD_3)_2SO$] δ 2.04 (s, 3, $COCH_3$), 2.13 (s, 6, $COCH_3$), 3.73 (s, 3, $NHCOCH_3$), 4.20–4.53 (m, 3, 4′-H, 5′-H), 5.40 (br s, 2, CH_2CON), 5.55 (m, 1, 3′-H), 5.78 (m, 1, 2′-H), 6.28 (d, J = 5 Hz, 1, 1′-H), 7.45 (br s, 2, NH_2, ex), 9.40 (s, 1, 8-H, ex).

B. 2′,3′,5′-Triacetylguanosine[12] (1.005 g, 2.46 mmol) and 2-bromoacetamide (0.683 g, 4.91 mmol) in anhydrous DMA (20 mL) was kept at room temperature for 12 days. TLC analysis of the reaction mixture showed a similar pattern of events to that described in part A. The solvent was evaporated in vacuo to a thick oil that was applied to a column of silica gel (80 g). Elution with a methanol-in-chloroform gradient (20 → 30%, v/v) gave the product as an oil which solidified on evaporation under high vacuum (0.67 g): ^1H NMR [$(CD_3)_2SO$] δ 2.01 (s, 3, $COCH_3$), 2.10 (s, 6, 2 × $COCH_3$), 4.23–4.61 (m, 3, 4′-H, 5′-H), 5.15 (br s, 2, CH_2CON), 5.51 (m, 1, 3′-H), 5.78 (m, 1, 2′-H), 6.23 (d, J = 5 Hz, 1, 1′-H), 7.43 (br s, ~1, NH, ex), 7.87 (br s, ~1, NH, ex), 9.47 (s, 1, 8-H, ex).

2-Chloroketene Diethyl Acetal (2). *Caution! 2-Chloroketene diethyl acetal is a mutagen and should be handled with caution in a well-ventilated hood, with suitable trapping.* In our hands, the published procedure[13] always led to a mixture of dichloroacetaldehyde diethyl acetal and 2-chloroketene diethyl acetal which was difficult to separate by distillation. We have found that the following modified procedure gives a good yield of a pure (^1H NMR) product. A solution of potassium *tert*-butoxide (21.0 g, 0.187 mol) in anhydrous *tert*-butyl alcohol (70 mL) was brought to gentle reflux, stirred magnetically, and dichloroacetaldehyde diethyl acetal (30.0 g, 0.160 mol) was added slowly by syringe over 10 min. The resulting mixture was stirred and heated under reflux under anhydrous conditions. The progress of the reaction was monitored by removal of an aliquot of the reaction mixture, dilution with CCl_4, and measurement of the ^1H NMR spectrum. When the pair of doublets due to the starting material had disappeared (3 days), the reaction mixture was centrifuged to remove KCl. The supernatant liquid was distilled, first at atmospheric pressure to remove *tert*-butyl alcohol, then fractionally under reduced pressure. Compound **2** was obtained as a colorless liquid: 16.2 g (67%); bp 98–103 °C (45 torr) [lit.[13] bp 166 °C (732–740 torr)]; ^1H NMR (CCl_4) δ 1.1–1.4 (m, 6, 2 × CH_3), 3.72 (q, J = 7 Hz, 2, OCH_2), 3.95 (q, J = 7 Hz, 2, OCH_2), 4.50 (s, 1, C=CH). Anal. Calcd for $C_6H_{11}ClO_2$: C, 47.84; H, 7.36; Cl, 23.54. Found: C, 48.14; H, 7.68; Cl, 23.50.

2-Ethoxy-5,6-dihydro-5-oxo-6-*n*-propylimidazo[1,2-*c*]pyrimidine (15). A solution if 2-chloroketene diethyl acetal (2.687 g, 17.8 mmol) and 1-*n*-propylcytosine[14] (0.682 g, 4.45 mmol) in anhydrous DMF (25 mL) was heated with magnetic stirring for 17 h at 80 °C (bath temperature). Anhydrous conditions were maintained throughout. The reaction was monitored by TLC (system B), which revealed the progressive disappearance of the starting material (R_f 0.08) and the formation of the product (R_f 0.60). Most of the DMF was evaporated in vacuo (bath temperature 50 °C) and the dark residue was applied to a column of silica gel (70 g). Elution with a methanol–chloroform gradient (0 → 4%, v/v) followed by pooling and evaporation of the appropriate fractions gave **15** as a pale-yellow solid, 0.663 g. Recrystallization from cyclohexane gave colorless needles (0.556 g, 56%): mp 93 °C; ^1H NMR ($CDCl_3$, 90 MHz) δ 0.99 (t, J = 8 Hz, 3, $CH_2CH_2CH_3$), 1.45 (t, J = 7 Hz, 3, OCH_2CH_3), 1.80 (m, 2, $CH_2CH_2CH_3$), 3.90 (t, J = 8 Hz, 2, $CH_2CH_2CH_3$), 4.14 (q, J = 7 Hz, 2, OCH_2CH_3), 6.43 (d, J = 7 Hz, 1, 8-H), 6.90 (s, 0.5, part of

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doublet of 7-H), 6.99 (s, 1.5, part of doublet of 7-H plus 3-H); low-resolution mass spectrum (EI, 70 eV) 221 (M^+, 72), 151 (100), 96 (14), 95 (33), 68 (11), 57 (11), 43 (25), 41 (25); IR (KBr) 3140, 3054, 2950 (br), 1699, 1630, 1570, 1425, 1300, 1055 cm^{-1}; UV (MeOH) λ_{max} 227 (ϵ 10600), 290 (ϵ 6080) nm, λ_{min} 243 (ϵ 2050) nm. Anal. Calcd for $C_{11}H_{15}N_3O_2$: C, 59.71; H, 6.83; N, 18.99. Found: C, 59.84; H, 6.87; N, 19.07.

4-*N*-(Ethoxycarbonyl)-1-*n*-propylcytosine (17). To a cooled (ice–water bath), stirred solution of compound **15** (0.100 g, 0.452 mmol) in dichloromethane (4 mL) was added purified MCPBA[15] (0.0859 g, 0.498 mmol). After 2.5 h at this temperature a further 1.1 equiv of MCPBA was added and the reaction was continued for another 1 h, when TLC (system B) indicated that no starting material was present. The reaction mixture was diluted with dichloromethane (35 mL), extracted with 1 M aqueous sodium metabisulfite solution (35 mL), dried ($MgSO_4$), and evaporated to give a pale-yellow oil (0.082 g). Purification by chromatography on silica gel (5 g) with an ether–chloroform gradient (0 → 15%, v/v) eluted the main product as a colorless oil which crystallized slowly, (0.030 g, 30%). Recrystallization from benzene–cyclohexane yielded colorless crystals: mp 133–134 °C dec; ^1H NMR ($CDCl_3$, 90 MHz) δ 0.99 (t, J = 8 Hz, 3, $CH_2CH_2CH_3$), 1.30 (t, J = 7 Hz, 3, OCH_2CH_3), 1.77 (m, 2, $CH_2CH_2CH_3$), 3.80 (t, J = 8 Hz, 2, $CH_2CH_2CH_3$), 4.21 (q, J = 7 Hz, 2, OCH_2CH_3), 7.14 (br d, J = 8 Hz, 1, 5-H), 7.50 (d, J = 8 Hz, 1, 6-H); high-resolution MS (EI, 70 eV) obsd 225.11098, $C_{12}H_{15}N_3O_3$, 225.1114; low-resolution MS (EI, 70 eV) 225 (M^+, 43), 183 (43), 180 (42), 153 (49), 138 (60), 124 (23), 111 (91), 81 (100), 43 (67), 41 (67). Anal. Calcd for $C_{10}H_{15}N_3O_3$: C, 53.32; H, 6.71; N, 18.66. Found: C, 53.19; H, 6.68; N, 18.82.

2-Ethoxy-5,6-dihydro-5-oxo-6-(2′,3′,5′-tri-*O*-acetyl-β-D-ribofuranosyl)imidazo[1,2-*c*]pyrimidine (12). A solution of 2′,3′,5′-tri-*O*-acetylcytidine (0.72 g, 1.95 mmol) and 2-chloroketene diethyl acetal (1.208 g, 8.00 mmol) in anhydrous DMF (5 mL) was stirred under anhydrous conditions at 80 °C (bath temperature) for 8 h. After this time no starting material could be detected by TLC (system B). Most of the DMF was removed in vacuo (bath temperature 50 °C) and the dark residue was dissolved in $CHCl_3$ and applied to a column of silica gel (70 g). Elution with a methanol in chloroform gradient (0 → 2%, v/v) gave **12** as a TLC homogeneous oil (0.728 g, 85%); R_f (system B) 0.65, R_f (tri-*O*-acetylcytidine) 0.29; high-resolution MS (FAB) obsd 438.1517, $C_{19}H_{24}N_3O_9$, 438.1513; low-resolution MS (FAB) 438 (MH^+, 100), 259 (37), 180 (83), 139 (40), 119 (38); ^1H NMR ($CDCl_3$, 90 MHz) δ 1.40 (t, J = 8 Hz, 3, OCH_2CH_3), 2.15 (m, 1, $COCH_3$), 4.10 (q, J = 8 Hz, 2, OCH_2CH_3), 4.33 (m, 3, 4′-H, 5′-H), 5.40 (m, 2, 2′-H, 3′-H), 6.20 (d, J = 6 Hz, 1, 1′-H), 6.49 (d, J = 8 Hz, 1, 8-H), 6.94 (s, 1, 3-H), 7.20 (d, J = 8 Hz, 1, 7-H).

N^4-(Ethoxy-2-chloroethylidene)-2′,3′,5′-tri-*O*-acetylcytidine (13). A solution of 2′,3′,5′-tri-*O*-acetylcytidine (0.369 g, 1.0 mmol) and 2-chloroketene diethyl acetal (0.604 g, 4.0 mmol) in anhydrous acetonitrile (4 mL) was kept at room temperature under anhydrous conditions. A progressive disappearance of the setting material to give a substance with much higher R_f was observed by TLC analysis of the reaction mixture. After 26 h, when no starting material remained, the reaction mixture was evaporated in vacuo and the residue was purified by chromatography on silica gel (20 g). Elution with chloroform gave unreacted 2-chloroketene diethyl acetal and then with chloroform–methanol (0 → 2%, v/v) gave **13** as a colorless oil (0.336 g, 71%) which was found to consist of two close-running components on TLC, the minor component of co-chromatographed cochromatographed with compound **12**. When this mixture was dissolved in dry DMF it was converted cleanly (80 °C/8 h) to the cyclized compound **12** (82%).

N^6-(1-Ethoxy-2-chloroethylidene)-2′,3′,5′-tri-*O*-acetyladenosine (14). A solution of 2′,3′,5′-tri-*O*-acetyladenosine (1.179 g, 3.0 mmol) and 2-chloroketene diethyl acetal (1.812 g, 12.0 mmol) in anhydrous DMF (5 mL) was stirred and heated under anhydrous conditions at 80 °C (bath temperature) for 2.7 h. After this time TLC (system B) indicated that most of the starting material had been converted to the imidate. Some of the highly fluorescent tricyclic compound **11** could also be detected by TLC as having an R_f roughly intermediate between product and starting material. Most of the DMF was removed in vacuo (bath temperature ca. 45 °C), and the orange residue was dissolved in a minimum amount of chloroform and applied to a column of silica gel (70 g). The column was eluted thoroughly with chloroform to remove excess 2-chloroketene diethyl acetal and then with a methanol–chloroform gradient until the product was eluted, typically in 1–2% (v/v) methanol in chloroform. The appropriate fractions were pooled and evaporated to give a colorless oil that was evacuated under high vacuum overnight to remove any DMF which had coeluted with the product: yield 0.994 g (67%); R_f (system B) 0.69, R_f (tri-*O*-acetyladenosine, system B) 0.44. The substance must be stored in vacuo over P_2O_5 if hydrolysis to tri-*O*-acetyladenosine is to be avoided: ^1H NMR (90 MHz, $CDCl_3$) δ 1.40 (t, J = 7 Hz, 3, OCH_2CH_3), 2.06, 2.09, 2.12 (3 s, 9, $COCH_3$), 4.14 (s, 2, CH_2Cl), 4.36–4.42 (m, 3, 4′-H, 5′-H's), 4.46 (q, J = 7 Hz, 2, OCH_2), 5.66 (m, 1, 3′-H), 5.94 (m, 1, 2′-H), 6.21 (d, J = 5 Hz, 1, 1′-H), 8.12 (s, 1, 8-H), 8.70 (s, 1, 2-H); low-resolution MS (FAB) 498 (MH^+). The ^1H NMR spectrum of **14** measured in $CDCl_3$ or $(CD_3)SO$ solution was indicative of only one compound.

8-Ethoxy-3-(2′,3′,5′-tri-*O*-acetylribofuranosyl)imidazo[2,1-*i*]purine (11). Compound **14** (0.250 g, 0.502 mmol) was dissolved in anhydrous DMF (5 mL) and heated with stirring at 80 °C (bath temperature) for 8 h under strictly anhydrous conditions. TLC (system B) after this time showed that the starting material had disappeared to give **11** and some tri-*O*-acetyladenosine had also been formed. The DMF was evaporated in vacuo and the residue was purified by chromatography on silica gel (60 g) by using gradient elution with methanol in chloroform (0 → 3%, v/v). The appropriate fractions were pooled and evaporated to give **11** as an oil homogeneous by TLC (0.090 g, 39%): ^1H NMR (90 MHz, $CDCl_3$) δ 1.43 (t, J = 7 Hz, 3, OCH_2CH_3), 2.14 (m, 9, $COCH_3$), 4.40 (m, 5, 4′-H, 5′-H, OCH_2CH_3), 5.63 (m, 1, 3′-H), 6.01 (m, 1, 2′-H), 6.18 (d, J = 5 Hz, 1′-H), 6.98 (s, 1, 7-H), 8.05 (s, 1, 2-H), 8.65 (s, 1, 5-H); high-resolution MS (FAB) obsd 462.1621, $C_{20}H_{24}N_4O_8$, 462.1625; low-resolution MS (FD) 461 (M^+, 100), 393 (28), 260 (9), 259 (9), 220 (8), 217 (9), 201 (3), 178 (19), 154 (6), 134 (7), 114 (13), 83 (28); R_f (system B) 0.55.

N^6-(Ethoxycarbonyl)-3-(2′,3′,5′-tri-*O*-acetylribofuranosyl)adenosine (16). To a cooled (ice–water bath), stirred solution of compound **11** (0.390 g, 0.846 mmol) in dichloromethane (10 ml) was added purified MCPBA (0.438 g, 2.538 mmol) in portions over 1 h. After the addition was complete, TLC examination of the reaction mixture (system B) revealed that no starting material remained and that two new components were present, R_f 0.67 and 0.42. The component with R_f 0.42 co-chromatographed with tri-*O*-acetyladenosine. The reaction mixture was diluted with chloroform (50 mL), extracted with 1 M aqueous sodium metabisulfite solution (50 mL) followed by saturated aqueous sodium bicarbonate solution (50 mL), dried ($MgSO_4$), and evaporated in vacuo to a pale-yellow oil (0.450 g). Further purification was affected by chromatography on silica gel (30 g) with a methanol–chloroform (0 → 2%, v/v) gradient as eluant. The appropriate fractions were pooled and evaporated to give a pale-yellow oil (0.203 g, 52%): ^1H NMR [$(CD_3)_2SO$], 90 MHz, δ 1.27 (t, J = 7 Hz, 3, $OCHCH_3$), 2.03, 2.06, 2.14 (3 s, 9, $COCH_3$), 4.17 (q, J = 7 Hz, 2, OCH_2CH_3), 4.25–4.55 (m, 3, 4′-H, 5′-H), 5.60 (m, 1, 3′-H),

Table I. ^1H Chemical Shifts of Compounds 7–11[a]

		8-H	2-H	N-H	1′-H
7	A	7.92	8.31		6.18 (m)
	B	8.37	8.19	7.3 (ex)	6.2 (m)

		2-H	5-H	7-H	N-H	1′-H
10	A	no data				
	B	8.55	9.25	8.17	10.87 (ex)	6.01 (d) J ~ 5 Hz
8	A	8.09	8.68	7.80 (br)	7.61 (ex)	6.27 (d) J ~ 5 Hz
	B	8.48	9.24	7.90	10.08 (ex)	6.31 (d) J ~ 5 Hz
9	A	8.01	8.53	6.89 (ex)		6.15 (m)
	B	8.43	9.02	7.05 (ex)	5.24 (br, ex)	6.30 (d) J ~ 5 Hz
11	A	8.05	8.65	6.98		6.18 (d) J ~ 5 Hz
	B	8.48	9.14	7.39		6.30 (d) J ~ 5 Hz

[a] In parts per million downfield from Me_4Si. All these data have been obtained specifically for this paper. A obtained in $CDCl_3$ solution. B obtained in $(CD_3)_2SO$ solution.

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Table II. ^{1}H Chemical Shifts of Compounds 3–6, 12[a]

		5-H	6-H		N-H	1′-H	Δδ
3	A	5.99 (d) $J \sim 8$ Hz	7.37 (d) $J \sim 8$ Hz		8.35 (v br)	5.88 (d) $J \sim 5$ Hz	1.38
	B	5.78 (d)	7.60 (d)		7.37 (b br)	5.80 (d)	1.82
		8-H	7-H	3-H	N-H	1′-H	Δδ
6a	A	6.52 (d) $J = 7.9$ Hz	7.24 (d) $J = 7.9$ Hz	8.08	9.23 (br)	6.30 (d) $J = 5.3$ Hz	0.72
	B	no data					
6b	A	no data					
	B	6.62 (d) $J = 7.8$ Hz	7.76 (d) $J = 7.8$ Hz	7.74	10.68 (ex)	6.02 (d) $J = 5.1$ Hz	1.14
4	A	6.69 (d) $J \sim 8$ Hz	7.18 (d) $J \sim 8$ Hz	7.73 (br)	9.10 (br)	6.29 (m)	0.49
	B	6.67 (d) $J \sim 8$ Hz	7.52 (d) $J \sim 8$ Hz	7.45	9.92 (ex)	6.16 (d) $J \sim 5$ Hz	0.85
5	A	6.49 (d) $J \sim 8$ Hz	7.15 (d) $J \sim 8$ Hz	6.90 (ex)	3.35 (ex)	6.25 (d) $J \sim 5$ Hz	0.66
	B	6.40 (d) $J \sim 8$ Hz	7.40 (d) $J \sim 8$ Hz	6.62 (ex)	5.04 (m, ex)	6.10 (d) $J \sim 5$ Hz	1.00
12	A	6.49 (d) $J \sim 8$ Hz	7.20 (d) $J \sim 8$ Hz	6.94		6.20 (d) $J \sim 5$ Hz	0.71
	B	6.65 (d) $J \sim 8$ Hz	7.54 (d) $J \sim 8$ Hz	7.10		6.13 (d) $J \sim 5$ Hz	0.89

[a] In parts per million downfield from Me_4Si. All these data have been obtained specifically for this paper. A obtained in $CDCl_3$ solution. B obtained in $(CD_3)_2SO$ solution.

5.97 (m, 1, 2′-H), 6.25 (d, $J = 5$ Hz, 1, 1′-H), 8.58 (s, 2, 2-H and 8-H), 10.4 (br s, 1, NH). The ^{1}H NMR measured in $CDCl_3$ showed singlets at δ 8.21 and 8.75 (2-H plus 8-H). Low-resolution mass spectrum (FAB) 466 (MH^+, 39), 259 (32), 208 (100), 162 (18), 139 (82), 136 (43), 119 (8); high-resolution mass spectrum (FAB) obsd 466.1576, $C_{19}H_{24}N_5O_9$, 466.1575.

N^6-[[(*tert*-Butoxycarbonyl)amino]carbonyl]-2′,3′,5′-tri-*O*-acetyladenosine (18). To a cooled (ice–water bath), stirred solution of compound 8 (0.390 g, 0.733 mmol) in chloroform (5 mL) was added in portions MCPBA (0.127 g, 0.733 mmol) and the resulting mixture was allowed to warm to room temperature over 3 h, when the reaction mixture was recooled and a further portion of MCPBA (0.171 g, 0.99 mmol) was added. The cooling bath was removed and the mixture allowed to warm to room temperature. The solution was diluted with chloroform (50 mL) and extracted in sequence with 1 M sodium metabisulfite solution (2 × 50 mL), saturated sodium bicarbonate solution 2 × 50 mL), and water (50 mL). The solution was then dried ($MgSO_4$) and evaporated in vacuo to give a pale-yellow oil that was purified by column chromatography (silica gel, 20 g) with a methanol–chloroform gradient (0 → 2%, v/v). Evaporation of the appropriate fractions gave compound 18 (0.118 g, 30%) as a foam. ^{1}H NMR (90 MHz, $CDCl_3$) δ 1.60 (s, 9, *t*-Bu), 2.08, 2.12 and 2.17 (3 s, 9, $COCH_3$), 4.41 (m, 3, 4′-H, 5′-H's), 5.80 (m, 1, 3′-H), 6.0–6.26 (m, 2, 2′-H, 1′-H), 8.55 (s, 1, 2-H or 8-H), 8.66 (s, 1, 2-H or 8-H), 10.7 (br s, 1, NH), 11.6 (br s, 1, NH); IR (film) 3150 (br), 2950, 1795, 1750, 1670, 1625, 1530, 1460, 1375, 1225 (br), 1140, 1080, 740 cm^{21}; high-resolution MS (FAB) obsd 537.1954, $C_{22}H_{29}N_6O_{10}$, 537.1945; low-resolution MS (FAB) 537 (MH^+, 30), 481 (14), 394 (5), 279 (18), 259 (47), 223 (45), 205 (12), 179 (36), 162 (23), 139 (100), 136 (71).

N^2-(1-Ethoxy-2-chloroethylidene)-2′,3′,5′-tri-*O*-acetylguanosine. A solution of 2′,3′,5′-tri-*O*-acetylguanosine (1.228 g, 3.0 mmol) and 2-chloroketene diethyl acetal (1.812 g, 12.0 mmol) in dry DMF (10 mL) was stirred magnetically and heated at 75–80 °C (bath temperature) for 15 h under anhydrous conditions. TLC (system C) after this time revealed that only a trace of starting material was present and a new substance of higher R_f had formed. Most of the DMF was evaporated in vacuo, and the residue was purified by chromatography on silica gel (75 g). Elution with a methanol–chloroform gradient (0 → 5%, v/v) first gave unreacted 2-chloroketene diethyl acetal, then a small amount of a fluorescent substance followed by the product. The appropriate fractions were pooled and evaporated to give N^2-(1-ethoxy-2-chloroethylidene)-2′,3′,5′-tri-*O*-acetylguanosine as a TLC homogeneous violet foam (1.337 g, 74%): R_f (system C) 0.65, R_f (tri-*O*-acetylguanosine) 0.45; ^{1}H NMR ($CDCl_3$, 90 MHz) δ 1.40 (t, J = 8 Hz, 3, OCH_2CH_3), 2.10 (m, 9, $COCH_3$), 4.23–4.54 [m, 7, OCH_2CH_3, 5′-H's, 4′-H containing a singlet at 4.40 (CH_2Cl)], 5.50 (m, 1, 3′-H), 5.75 (m, 1, 2′-H), 6.03 (d, $J = 5$ Hz, 1, 1′-H), 7.88 (s, 1, 8-H), 11.69 (br s, 1, NH); low-resolution MS (FAB) 514 (MH^+, 21), 480 (14), 436 (10), 410 (10), 259 (89), 222 (18), 139 (100), 115 (8).

[8-^{2}H]-2′,3′,5′-Tri-*O*-acetyladenosine ([8-^{2}H]7). 2′,3′,5′-Tri-*O*-acetyladenosine (2.0 g, 5.09 mmol) was dissolved in a minimum amount of boiling C_2H_5OD (~25 ml) and D_2O (~10 mL) was added to the boiling solution in portions so that homogeneity was maintained and the mixture was stirred and heated under reflux (N_2). The extent of deuteriation was monitored by removing aliquots of the reaction mixture, dilution with $CDCl_3$, and measurement of the ^{1}H NMR spectrum. When the signal at δ 7.92 had diminished to ca. 5% of its original intensity (50 h) the reaction mixture was cooled to room temperature, when crystals (1.65 g, 82.5%) were deposited. Evaporation of the filtrate yielded more product (0.25 g).

Preparation of [2-^{2}H]8. This substance was prepared from [8-^{2}H]-2′,3′,5′-tri-*O*-acetyladenosine (0.393 g, 1.0 mmol) and *N*-(*tert*-butoxycarbonyl)-2-bromoacetamide (0.238 g, 1.0 mmol) as described for the nondeuterated compound: yield 0.166 g (31%).

Preparation of [2,7-$^2H_2$]8. A sample of [2-^{2}H]8 (0.05 g, 0.094 mmol) was dissolved in $(CD_3)_2SO$ (0.5 mL) in an NMR tube and D_2O (0.1 mL) was added. The tube was heated at 85–90 °C (bath temperature) for 6 h. The solution was cooled to room temperature and the ^{1}H NMR spectrum measured.

^{1}H NMR Assignments. The heterocycle ^{1}H NMR signals of the adenosine-derived compounds (Table I) were assigned unambiguously by deuterium labeling experiments. Each of these compounds shows three discrete singlet resonances in the low-field region of the spectrum. Two of these resonances (those attached to a five-membered ring) should be susceptible to substitution by deuterium. There are three reasons why this should be so: A. It is known that the C8 imidazole ring proton of adenosine undergoes substitution with deuterium when heated in D_2O solution.[16] B. It is known that the 3-position of imidazo[1,2-*a*]-pyridines is susceptible to electrophilic substitution.[17] C. An amino–imino type tautomerism possible in ring C makes it likely that the proton attached to C7 would undergo deuterium exchange. That deuterium exchange does, in fact, occur at the C2 and C7 positions in compounds **8**, **9**, **10**, and **11** will become obvious from the following discussion.

When compound **8** was treated with boiling C_2H_5OD/D_2O solution for 16 h and the ^{1} H NMR spectrum was obtained in $(CD_3)_2SO$, it was found that the signal at δ 9.24 remained undiminished in intensity, whereas the signals at δ 8.48 and 7.90 became, respectively, ca. 20 and 5% relatively as intense. The signal at δ 9.24 is therefore assigned to the C5 proton in **8**. The assignment was completed by synthesizing [2-^{2}H]8 in the normal manner from [8-^{2}H]-2′,3′,5′-tri-*O*-acetyladenosine ([8-^{2}H]**7**) and $BrCH_2CONHCO_2Bu$-*t*. The ^{1}H NMR spectrum of this substance in $(CD_3)_2SO$ solution showed that the resonance at δ 8.48 was ca. 5% of its relative intensity in the spectrum of **8**. The signal at δ 8.48 is therefore due to the proton at C2 and that at δ 7.90 is due to the proton at C7. Furthermore, when [2-^{2}H]8 was treated at 85–90 °C in $(CD_3)_2SO/D_2O$ solution for 6 h the resonance at δ 7.90 diminished to ca. 1% of its original intensity, indicating conversion of [2-^{2}H]**8** to [2,7-$^2H_2$]**8**. The overall process is summarized in Scheme III.

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A similar sequence of isotopic labeling experiments allowed the resonances at δ 9.25, 8.55, and 8.17 to be assigned to the protons at C5, C2, and C7, respectively, in compound **10**. When [2-^{2}H]**10**, synthesized in the usual manner from [8-^{2}H]**7** and $BrCH_2CONHCOCH_3$ was treated at 85–90 °C with $(CD_3)_2SO/D_2O$ for 12 h, the resonance at δ 8.17 diminished to ca. 10% of its intensity in the unlabeled case.

The ^{1}H NMR spectrum of compound **9** measured in dry $(CD_3)_2SO$ solution showed three discrete resonances at δ 9.02, 8.43, and 7.05. Upon addition of 2–3 drops of D_2O to the solution, the signal at δ 7.05 immediately diminished to ca. 5% of its original intensity. Such a rapid rate of exchange suggests a dynamic amino–imino tautomerism in the enamine-type system. The resonance at δ 7.05 is therefore due to the proton at the C7 position. When compound [2-^{2}H]**8** was converted to the free amine [2-^{2}H]**9** using TFA at room temperature, the ^{1}H NMR spectrum measured in $(CD_3)_2SO$ solution showed that the resonance at δ 8.43 for **9** was replaced with one of ca. 5% the intensity. This resonance is therefore due to the proton attached to C2 in **9**.

Compound **11** also showed three discrete singlet resonances at δ 9.14, 8.48, and 7.39 in the low-field region of the ^{1}H NMR spectrum measured in $(CD_3)_2SO$. In the ^{1}H NMR spectrum of [2-^{2}H]**11** (prepared from [8-^{2}H]-2′,3′,5′-tri-*O*-acetyladenosine ([8-^{2}H]**7**) and **2** in the normal manner), the resonance at δ 8.48 had ca. 5% of the intensity of the other singlet resonances. Furthermore, when 2–3 drops of D_2O were added to the $(CD_3)_2SO$ solution at room temperature, a progressive decrease in the intensity of the resonance at δ 7.39 indicated a conversion of [2-^{2}H]**11** to [2,7-^{2}H]**8** over several days. This sequence of deuterium labeling experiments leads to the unequivocal assignments: δ 9.14 (5-H), 8.48 (2-H), and 7.39 (7-H).

An interesting switch over in the relative rates of exchange of the labile protons in **8** and **10** is evident. in **8**, the 7-H exchanges first (16 h) followed by 2-H (24 h) in boiling C_2H_5OD/D_2O solution. In **10**, 2-H exchanges first (16 h) followed by 7-H (120 h). Such a large change in the lability of the 7-H in these compounds probably can be attributed to the electronic effect of the 8-substituent in the etheno bridge.

A general trend is apparent in the ^{1}H NMR spectra of compounds **8**, **9**, **10**, and **11** measured in $(CD_3)_2SO$ solution, namely, that *conversion of tri-O-acetyladenosine into an 8-substituted imidazo[2,1-i]purine causes a marked (0.83–1.06 ppm) downfield shift of the 2-proton (adenosine numbering).* A similar effect has been observed when adenosine is converted to 1,N^6-ethenoadenosine hydrochloride (εAdo·HCl), but because a different solvent (D_2O) was used for measurement of the ^{1}H NMR spectrum a direct parallel cannot be drawn.[15]

No deuterium labeling was required for the unambiguous assignment (see Table II) of the ^{1}H NMR signals of the cytidine-derived compounds, since chemical shift data combined with signal multiplicity gave sufficient information. From the data assembled in Table II it is apparent that annelation of a five-membered ring onto the cytidine nucleus causes a marked downfield shift of 0.50–0.70 ppm ($CDCl_3$) or 0.62–0.89 ppm [$(CD_3)_2SO$] for the 5-H (cytidine numbering) with a concomitant upfield shift of 0.13–0.22 ppm ($CDCl_3$) or 0.06–0.20 ppm [$(CD_3)_2SO$] for the 6-H. Compound **6b** is anomalous in this respect since the ^{1}H NMR spectrum measured in $(CD_3)_2SO$ solution shows a 0.16 ppm downfield shift of the original 6-H relative to tri-*O*-acetylcytidine. This is probably due to the fact that the ribose hydroxyl functions are unprotected since the data for its parent tetraacetyl derivative **6a** measured in $CDCl_3$ are in line with the general trend described. *The overall effect of annelation on the cytidine H-5/H-6 spin system is to decrease markedly the chemical shift difference (Δδ) between the pyrimidine ring protons.* This feature of the ^{1}H NMR data is characteristic for this series of compounds.

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Registry No. **1a**, 96394-42-2; **1b**, 81836-87-5; **1c**, 96394-43-3; **2**, 42520-09-2; **3**, 56787-28-1; **4**, 96394-44-4; **5**, 96394-45-5; **6a**, 96394-46-6; **6b**, 61671-71-4; **7**, 7387-57-7; [8-^{2}H]-**7**, 96394-47-7; **8**, 96394-48-8; [2-^{2}H]-**8**, 96394-49-9; [2,7-$^2H_2$]-**8**, 96394-50-2; **9**, 96394-51-3; **10**, 96394-52-4; **11**, 96394-53-5; **12**, 96394-54-6; **13**, 96394-55-7; **14**, 96394-56-8; **15**, 96394-57-9; **16**, 33422-70-7; **17**, 96394-58-0; **18**, 96394-59-1; 2-bromoacetamide, 683-57-8; 2-bromoacetyl bromide, 598-21-0; *N*-(bromoacetyl)acetamide, 34002-90-9; 8-(*N*-acetylamino)-3-(2′,3′,5′-tri-*O*-acetyl-β-D-ribofuranosyl)imidazo[2,1-*i*]purine, 96394-60-4; 2′,3′,5′-tri-*O*-acetylguanosine, 6979-94-8; 6-(*N*-acetylamino)-3-(2′,3′,5′-tri-*O*-acetyl-β-D-ribofuranosyl)-5,9-dihydroimidazo[1,2-*a*]purin-9(3*H*)-one, 96394-61-5; 7-(*N*-acetylamino)-1-(2′,3′,5′-tri-*O*-acetyl-β-D-ribofuranosyl)-4,6-dihydroimidazo[2,1-*b*]purin-4(1*H*)-one, 96394-62-6; 7-(aminocarbonylmethyl)-8-bromo-7,8-dihydro-2′,3′,5′-tri-*O*-acetylguanosine, 96394-63-7; dichloroacetaldehyde diethyl acetal, 619-33-0; 1-propylcytosine, 22919-46-6; N^2-(1-ethoxy-2-chloroethylidene)-2′,3′,5′-tri-*O*-acetylguanosine, 96394-64-8.