white resin, bp 170 °C/0.04 mmHg: HRMS, calcd for $C_{22}H_{30}O_2$ 326.225, found 326.224; ^{1}H NMR ($CDCl_3$) δ 0.89 (t, 3 H, J = 6.8 Hz, ϵCH_3), 1.09 (s, 3 H, 6α-Me), 1.36 (s, 3 H, 6β-Me), 1.91 (d, 3 H, J = 1.1 Hz, 9-Me), 2.49 (t, 2 H, αCH_2), 3.18 (d, 1 H, J = 11.0 Hz, 10a-H), 4.84 (s, 1 H, =CH), 4.99 (s, 1 H, OH, D_2O exchangeable), 5.09 (s, 1 H, =CH′), 5.68 (br d, 1 H, J = 3.4 Hz, 8-H), 6.35 (d, 1 H, J = 1.4 Hz, ArH), 6.38 (d, 1 H, J = 1.5 Hz, ArH′); IR (CH_2Cl_2) 3510 cm^{-1} (OH). Anal. Calcd for $C_{22}H_{30}O_2$: C, 80.94; H, 9.26. Found: C, 80.71; H, 9.31.

(5a*R*-*trans*)-5a,7,9,10c-Tetrahydro-5,5,8-trimethyl-2-pentyl-5*H*,6*H*-[2]benzopyrano[5,4,3-*cde*][1]benzopyran (12) [*O*,10-Methano-Δ^9-THC]. 10-(Hydroxymethyl)-Δ^9-THC (3.1 g, 9.0 mmol) in dry pyridine (50 mL) was treated with *p*-toluenesulfonyl chloride (5.14 g, 27.0 mmol) in dry pyridine (10 mL) over a period of 0.5 min with stirring. After being stirred for 2.2 h, the reaction was quenched by the addition of H_2O (6 mL) and stirred for 10 min. Volatiles were evaporated in vacuo and the residue was dissolved in CH_2Cl_2 and extracted twice with saturated $NaHCO_3$, 1 N HCl, and H_2O. Drying over Na_2SO_4 and removing the solvent in vacuo afforded a foam that was eluted from silica gel (30 g) with CH_2Cl_2. The product-containing fractions were combined and rechromatographed from a Merck size C silica gel column, eluting with 20% CH_2Cl_2/hexane, to afford 534 mg (18%) of the title compound followed by 338 mg (12%) of 10-methylene-Δ^8-THC. The title compound was distilled bulb-to-bulb to afford a pale yellow resin, bp 175 °C/ 0.04 mmHg. On exposure to air, this material yellowed and showed degradation products by HPLC (Waters RCM C-18, 85% CH_3CN/H_2O, 280 nm): HRMS, calcd for $C_{22}H_{30}O_2$ 326.225, found 326.225; ^{1}H NMR ($CDCl_3$) δ 0.87 (t, 3 H, J = 6.7 Hz, ϵCH_3), 1.26 (s, 3 H, 6α-Me), 1.42 (s, 3 H, 6β-Me), 1.69 (d, 3 H, J = 0.88 Hz, 9-Me), 2.17 (m, 2 H, 8-CH_2), 2.45 (t, 2 H, J = 7.6 Hz, αCH_2), 3.10 (br d, 1 H, J = 9.6 Hz, 10a-H), 4.52 (br d, 1 H, J = 13.2 Hz, *O*,10-CH), 4.90 (d, 1 H, J = 13.0 Hz, *O*,10-CH′), 6.20 (d, 1 H, J = 1.3 Hz, ArH), 6.23 (d, 1 H, J = 1.3 Hz, ArH′). Anal. Calcd for $C_{22}H_{30}O_2$: C, 80.94; H, 9.26. Found: C, 80.63; H, 9.07.

Cleavage of (β-Methoxyethoxy)methyl Phenyl Ether (5). (β-Methoxyethoxy)methyl phenyl ether (207 mg, 1.13 mmol) and allyltrimethylsilane (0.27 mL, 1.71 mmol) in 5 mL of dry CH_2Cl_2 were cooled to −20 °C under N_2 and treated with $TiCl_4$ (0.15 mL, 1.37 mmol) in five portions over 10 min with stirring. After being stirred for 1 h at −20 °C, the reaction was quenched with aqueous $NaHCO_3$ and extracted with ether (2×). The combined ether extracts were washed with H_2O (3×), dried over $MgSO_4$, and evaporated to a light yellow liquid (275 mg) after pumping at high vacuum. Elution from silica gel (12 g) with 9 to 11% CH_2Cl_2/hexane afforded 4-phenoxy-1-butene (36 mg, 22%) as the major and earliest eluting product: ^{1}H NMR, 90 MHz ($CDCl_3$) δ 2.60 (q, [dt], 2 H, J = 6 Hz, 3-CH_2), 4.10 (t, 2 H, J = 6.5 Hz, 0-CH_2), 5.20 (m, 2 H, =CH_2), 6.02 (m, 1 H, CH=), 7.01 (m, 3 H, ArH_3), 7.36 (m, 2 H, ArH_2). The compound was unstable to GC and MS at 80 °C.

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Registry No. 1, 5957-75-5; 2, 131323-38-1; 3, 131351-99-0; 4, 131323-39-2; 5, 131323-44-9; 6, 2653-89-6; 7, 1972-08-3; 8, 82459-42-5; 9, 131323-40-5; 10, 131323-41-6; 11, 131323-42-7; 12, 131323-43-8; $Br(CH_2)_3OH$, 627-18-9.

Supplementary Material Available: 2D heteronuclear correlation spectra of 3 and 12 (2 pages). Ordering information is given on any current masthead page.

Synthesis of 1- and 1,2,2′-Deuteriated Deoxyribose and Incorporation into Deoxyribonucleosides

Richard P. Hodge,† Charles K. Brush,‡ Constance M. Harris,* and Thomas M. Harris*

Department of Chemistry and Center in Molecular Toxicology, Vanderbilt University, Nashville, Tennessee 37235

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Thymidine, deoxycytidine, deoxyadenosine, and deoxyguanosine have been prepared with deuterium substitution at position 1′ and at positions 1′, 2′, and 2″ from deuteriated deoxyribose. The synthetic strategy involved reduction of the bis(*tert*-butyldimethylsilyl) derivative **4a** of 2-deoxyribonolactone (**3a**) with Dibal-D followed by deprotection (HCl/MeOH and tetrabutylammonium fluoride) to give 1-deuterio methyl glycoside **7a** which was converted to the 1-deuterio 3,5-ditoluoyl methyl glycoside **8a**. Preliminary exchange of 2-deoxyribonolactone with NaOMe/MeOD brought about 2,2′-dideuteriation; treatment as above gave the 1,2,2′-trideuterio 3,5-ditoluoyl methyl glycoside **8b**. **8a** and **8b** were condensed with heterocyclic bases via α-chloro derivatives **9a** and **9b** to form deoxynucleosides. New methods were utilized for preparation of deoxycytidine and deoxyguanosine which are improvements over published procedures.

The investigation of interactions of both small and large molecules with oligonucleotides as models of their interactions with genomic DNA is a rapidly expanding field of study[1] due to the ready availability of sequence-specific DNA oligomers via automated DNA synthesis[2] and to the development of highly sophisticated instrumentation and software for analysis of solution structure, particularly in the area of NMR spectroscopy.[3] Two-dimensional NMR correlated spectroscopy (COSY) allows the spectral assignment of protons connected via small numbers of bonds

† Present address: Milligen Biosearch, Division of Millipore, 186 Middlesex Turnpike, Burlington, MA 01803.

‡ Present address: Pharmacia-P. L. Biochemicals, 2202 N. Bartlett Ave., Milwaukee, WI 53202.

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while 2D nuclear Overhauser enhancement spectroscopy (NOESY) yields information about proximal protons which may not be bonded to one another at all. Application of the latter methodology has been important in the assignment of signals in double-stranded oligonucleotides where it is possible to follow NOEs down a strand from a base proton (purine H8 or pyrimidine H6) to the sugar to which the base is attached (H1′, H2′, and H2″) and to the H1′ of the adjacent 5′ sugar. The strategy is of particular value for spectral assignment of B-form DNA. Once the ^1H assignment of a particular DNA segment is known, the dynamic conformation and interactions (covalent or noncovalent) with other molecules can be studied. At present, the assignment strategy is limited to oligomers of no more than 12–15 base pairs. When instruments of higher field strength become available, it should become possible to extend existing methodology to larger oligomers. At present, however, methods such as spectral editing for resolving spectra and assigning signals will be required for the study of longer and more complex strands of DNA. Editing can be achieved instrumentally, for example, by three-dimensional NMR[4] or chemically by isotopic substitution. Suppression of nonessential proton resonances can be achieved by regiospecific incorporation of deuterium. We have recently published studies on reversible deuteriation of the bases as a tool for identifying aromatic resonances in a non-self-complementary dodecamer duplex[5a] and on the use of methyl-deuteriated thymidine for the assignment of individual residues in a TTTT tetramer.[5b]

By far the most serious problems with spectral congestion involve the deoxyribose signals, and it would be useful to apply specific deuteriation techniques to this portion of the DNA molecule. Of particular value in this regard would be deuteriation at the 1′, 2′, and 2″ positions because of their strategic involvement in the assignment methodology. One potential application would be in the study of conformations which cannot be achieved in short strands of DNA. Another would be in the study of adducts with carcinogens and drugs, where major conformational alterations can interfere with systematic assignment; deuterium substitution of remote residues in the oligonucleotide would allow easier ^1H assignment of regions of interest near the site of the adducted base. In a ^1H spectrum in which only selected sugar protons are visible the measurement of coupling constants would be facilitated, thus permitting determination of the conformations of furanose rings. Deuteriated oligonucleotides would also be useful for studies of molecular motion in the solid state, such as those recently described by Roy et al.[6]

The most extensive use of deuteriation for the study of nucleotides thus far reported is the work of Danyluk and his co-workers who prepared a wide range of ribonucleotides in which one residue was perdeuteriated.[7] The deuteriated mononucleotides, isolated from the alga *Synechoccus lividus* which was grown in D_2O, were coupled to the nondeuteriated residue either chemically or enzymatically. These workers were able to assign completely the ^1H NMR spectra of the dinucleotides and to establish conformations. Roy has also prepared 2′,2″-dideuteriothymidine and 2′,2″-dideuteriodeoxyguanosine by enzymatic processes.[6]

Most of the approaches to deuteriated deoxyribonucleosides, however, have been by chemical synthesis.[8] Some have been carried out at the glycoside level before coupling with heterocyclic bases; others have employed nucleosides themselves as starting materials. Fraser-Reid et al.[8h] synthesized 2-deuterio- and 2′-deuteriodeoxyriboses by a procedure involving stereospecific reduction of a 2,3-dehydrohexopyranose with $LiAlD_4$ and Lemieux–Johnson oxidation of the resulting glycal. Each deuteriodeoxyribose was then converted to the corresponding 2′-deuteriodeoxycytidine by the method of Fox.[8i,j] 2′-Deuteriodeoxyuridine and other 2′-deuteriodeoxynucleosides have been prepared similarly.[8k,l] Wong and Gray[8m] reduced the ketene dithioacetal derived from D-arabinose with $LiAlD_4$ to get stereoselective incorporation of deuterium at C-2 of deoxyribose. Quenching the reduction reaction with D_2O afforded the 1,2-dideuterio derivative.

H/D exchange using hydrogenation/dehydrogenation catalysts in D_2O has been widely used for incorporation of deuterium into sugars.[9] This method tends to be nonspecific, although there is definitely a structure-dependent difference in exchange rates;[9d] problems include epimerization and incomplete exchange. Tritium has been incorporated into the 2-position of deoxyribose by base-catalyzed exchange in ethanolic tritiated water.[10] Tritium has also been incorporated into the 1-position of ribose by reduction of ribonolactone with $NaBT_4$, but only a 10% yield was obtained.[11]

Recently Chattopadhyaya and co-workers published several papers on the synthesis of deoxyribose-deuteriated deoxynucleosides via deoxyribose derivatives.[12] They

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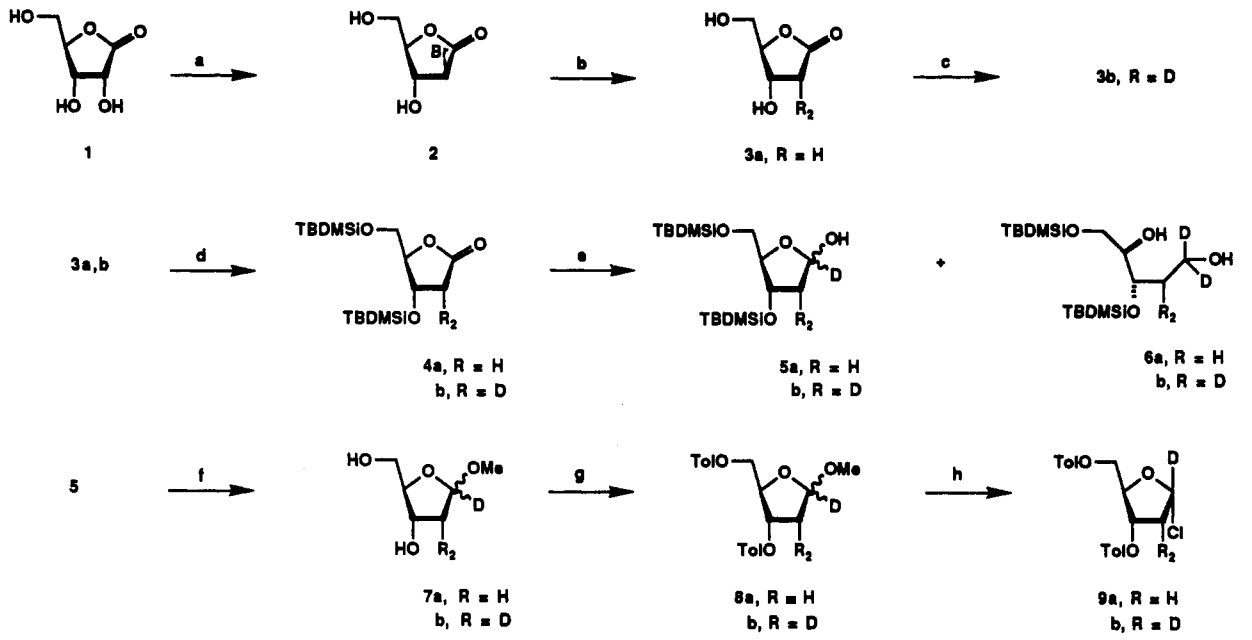
Scheme I[a]



[a] (a) HBr, HOAc; (b) n-Bu_3SnH; (c) NaOMe, MeOD; (d) $TBDMSiONO_2$ (e) Dibal-D; (f) 0.01% HCl, MeOH; (g) p-TolCl; (h) HCl, HOAc.

detailed a method of synthesizing stereospecifically all eight 2′- and 2″-deuterio-2′-deoxynucleosides by reductive opening of 2,3-anhydrofuranosides with $LiAlD_4$.[12a] Subsequently they reported[12b] the synthesis of 2′,2″-dideuterio-2′-deoxynucleosides by an oxidation–reduction sequence first described by Hansske et al.[13a] and Robins et al.[13b] This route starts with an aldose in the pyranose form and hence is not easily adapted to introduction of deuterium at the 1-position. A third paper described the synthesis of 2′,2″,3′,4′-tetradeuterio-2′-deoxynucleosides by a combination of Raney Ni deuteriation and deoxygenation at the 2′-position with tributyltin deuteride.[12c]

We now report new methodology for preparation of deoxynucleosides deuteriated at the 1′-, 2′-, and 2″-positions of deoxyribose. We considered three different strategies for the preparation of sugar-deuteriated deoxynucleosides. (1) Deuteriation at the nucleoside level. This would not permit deuteriation at the 1′-position; methodology using catalytic exchange as discussed above did not look promising. (2) Synthesis of deuteriated ribose followed by incorporation into ribonucleosides and deoxygenation at C-2′. Glycosylation proceeds more cleanly with ribose derivatives than with deoxyribose but the necessity of carrying out several more reactions rendered this strategy unappealing. (3) Preparation of deuteriated deoxyribose followed by nucleoside synthesis. In spite of potential problems with regio- and stereospecificity in the coupling reaction, we chose to pursue this route because of the facility with which deuterium could be introduced at positions 1 and 2 of deoxyribose.

Results and Discussion

Deuteriated Deoxyribose. In the present study 2-deoxyribonolactone (**3a**) was used as starting material for the synthesis of the deuteriated deoxynucleosides. It is readily prepared from ribonolactone which is commercially available;[14] a synthesis of **3a** from L-arabinose has also been described recently.[15] Whereas deoxyribose exists in an equilibrium mixture of pyranose and furanose structures and requires protection of the terminal hydroxyl group to keep it in the latter form, deoxyribonic acid shows preference for the γ-lactone over the δ. H/D exchange can be brought about on the lactone under basic conditions; deoxyribose also undergoes base-catalyzed exchange but is susceptible to aldol condensations. The lactone carbonyl can be reduced to the aldehyde using Dibal; deuterium can be placed at the C-1 position using Dibal-D.

Ribonolactone **1** was converted to the 2-bromo-2-deoxy lactone **2** by treatment with HBr in acetic acid.[16] Reductive dehalogenation of **2** by hydrogenation over palladium–carbon[17] gave poor yields of deoxyribonolactone (**3a**), but dehalogenation using tributyltin hydride was essentially quantitative.[17,18] The hydroxyl groups of **3a** were quantitatively protected as the *tert*-butyldimethylsilyl ethers by treatment with *tert*-butyldimethylsilyl nitrate to give **4a**.[19]

Lactone **4a** was reduced with Dibal-D[20] to give 86% of protected 1-deuteriodeoxyribose **5a**. Care had to be exercised to avoid overreduction. By carrying out the reaction at low temperature and high dilution the reaction was slow enough that it could be monitored by TLC and quenched with methanol at first sign of formation of diol **6a**. Small amounts of unreacted starting material **4a** (~

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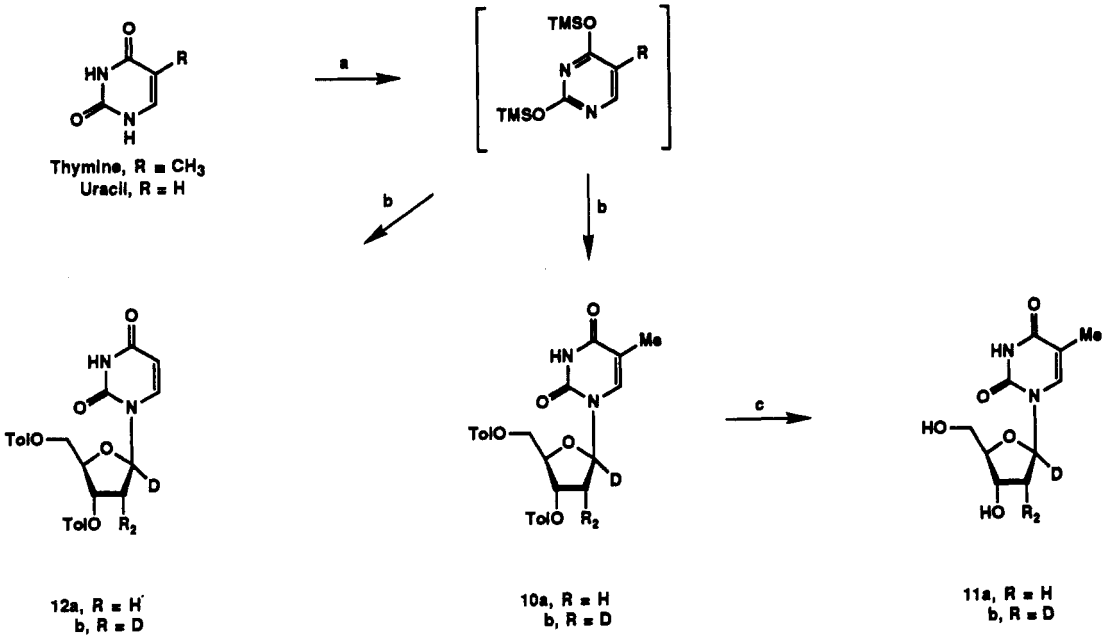
Scheme II[a]



12a, R = H
b, R = D

10a, R = H
b, R = D

11a, R = H
b, R = D

[a] (a) HMDS; (b) **9ab**, $CHCl_3$; (c) NH_3, MeOH.

10%) could be recovered. Yields using this method were typically 80–85% with deuterium incorporation greater than 99% as determined by ^1H NMR analysis. The problem with overreduction might be diminished by using diisoamylborane instead of Dibal.[21]

Treatment of **5a** with methanolic HCl gave the methyl glycoside as an anomeric mixture; the silyl groups were removed with tetrabutylammonium fluoride[22] to give **7a** in 90% yield. NMR analysis of the reaction mixture after methanolic HCl treatment indicated that loss of ~50% of the silyl protecting groups had already occurred. Only a trace of the methyl pyranoside could be detected, suggesting that acetal formation occurred significantly faster than deprotection of position 5. Compound **7a** was then converted to the 3,5-bis(toluoyl ester) (**8a**) in 95% yield using the acid chloride.[23]

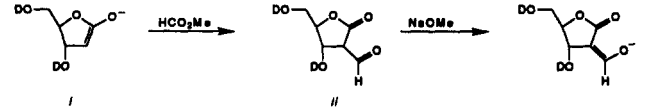
Synthesis of the 1,2,2′-trideuteriated species **8b** was carried out by the same route with inclusion of base-catalyzed H/D exchange at C-2 of **3a**. Exchange using sodium methoxide in CH_3OD occurred rapidly, but the extent of deuteriation was found to be highly dependent upon the purity of the CH_3OD. Material prepared by hydrolysis of trimethyl orthoformate with D_2O was found to cause gradual loss of sodium methoxide. Exchange would continue if additional sodium methoxide was added. The loss of sodium methoxide is ascribed to the presence of trace quantities of methyl formate which could undergo displacement to give dimethyl ether and sodium formate.[24] The problem was subsequently circumvented using CH_3OD prepared by hydrolysis of $(MeO)_4Si$ with D_2O. The exchange reaction was terminated by addition of CF_3CO_2D/D_2O. Deuteriated lactone **3b** was isolated in 85% yield with ~95% deuteriation at the 2-position (β-face) and ~85% deuteriation at the 2′-position; **3b** was converted to **8b** as described above.

This route to deuteriated deoxyribose complements those of Chattopadhyaya et al.[12] which make no provision for incorporation of deuteriation at C-1. The present approach for introduction of deuterium at positions 2,2′ is also more efficient than the one they described.[12b]

Deoxynucleoside Assembly. Nucleosides are assembled by condensation of the heterocyclic bases with the sugars. Numerous variations on this strategy exist involving different protection and activation for the base and carbohydrate, and in some cases transformations after formation of the nucleoside linkage.[25] The synthesis of 2′-deoxynucleosides has been troublesome because of difficulty of obtaining regio- and stereospecificity in the condensation reaction. One of the best intermediates was developed by Hoffer,[23] who found that one anomer (later shown to be the α-anomer[26]) of the 1-chloro 3,5-bis(toluoyl ester) of deoxyribose crystallizes preferentially and in high yield during treatment of the methyl glycoside with HCl/HOAc. Condensations with this α-anomer, if carried out properly, occur predominantly with inversion of configuration to give β-linked deoxynucleosides. The strategy involves use of conditions which will minimize chloride ion catalyzed equilibration of the α and β anomers of the chloro sugar. Hoffer's chloro sugar was employed in the current syntheses.

Deuteriated thymidines **10a** and **10b** were synthesized by condensation of chloro sugars **9a** and **9b** with 2,4-di-*O*-(trimethylsilyl)thymine, a strategy developed by Hubbard.[27] The reaction was carried out in $CHCl_3$; subsequent

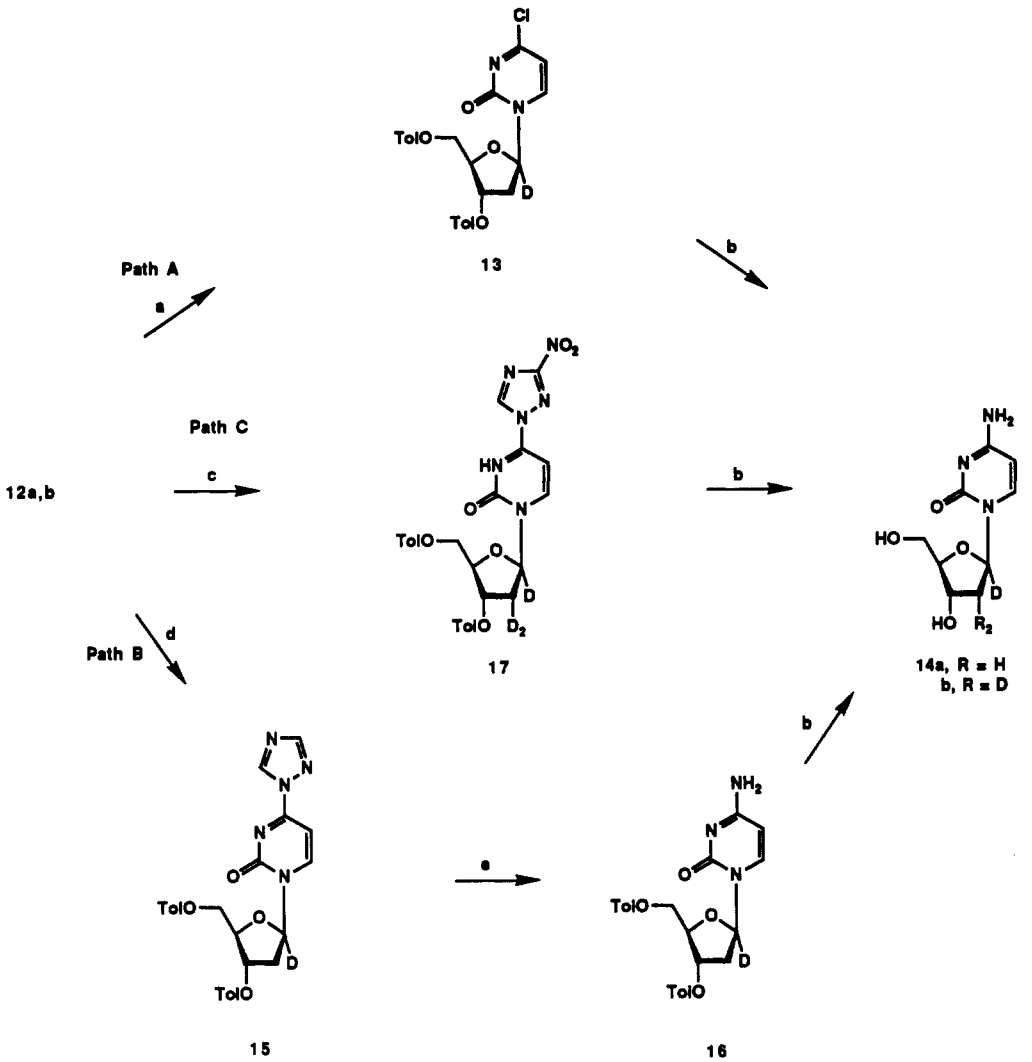
(21) Kohn, P.; Samaritano, R. H.; Lerner, L. M. *J. Am. Chem. Soc.* **1965**, *87*, 5475.
(22) Corey, E. J.; Venkateswarlu, A. *J. Am. Chem. Soc.* **1972**, *94*, 6190.
(23) (a) Hoffer, M. *Chem. Ber.* **1960**, *93*, 2777. (b) Hoffer, M.; Dushinsky, R.; Fox, J. J.; Yung, N. *J. Am. Chem. Soc.* **1959**, *81*, 4112.
(24) The loss of methoxide ion can be explained by acylation of anionic intermediate i by methyl formate to give the formylated product ii which could be reionized leading to consumption of methoxide.



i ii

(25) (a) Mizuno, Y. In *The Organic Chemistry of Nucleic Acids*; Elsevier: Amsterdam, 1986; Chapter 3. (b) Goodman, L. In *Basic Principles in Nucleic Acid Chemistry*; Ts'o, P. O. P., Ed.; Academic Press: New York, 1974; Chapter 2. (c) *Chemistry of Nucleosides and Nucleotides*, Townsend, L. B., Ed.; Plenum: New York, 1988.
(26) Bhattacharya, A. K.; Ness, R. K.; Fletcher, H. G., Jr. *J. Org. Chem.* **1963**, *28*, 428.

Scheme III[a]



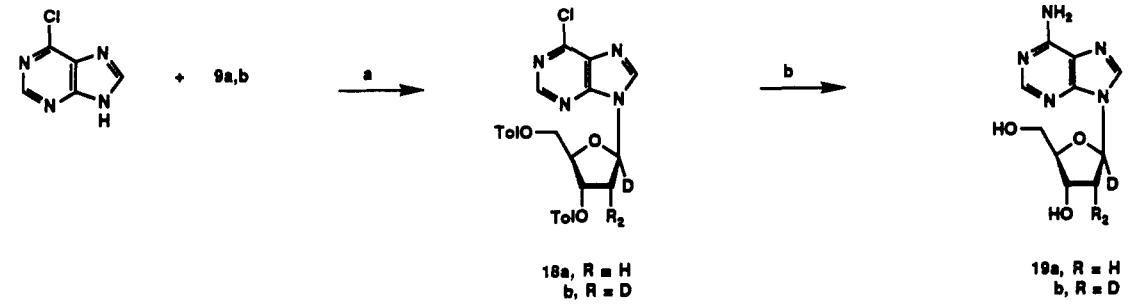
[a] (a) $SOCl_2$, DMF; (b) NH_3, MeOH; (c) 3-NO_2-triazole, Ph_2POCl; (d) triazole, $POCl_3$; (e) NH_4OH, dioxane.

deprotection with methanolic ammonia gave **11a** and **11b** in 91 and 81% yields, respectively.

The synthesis of 2′-deoxycytidine was not so straightforward. Chattopadhyaya, in his syntheses of deuteriated 2′-deoxycytidine,[12] employed cytosine under the Hubbard conditions. The reduced nucleophilicity of silylated cytosine compared to the bissilyl derivative of thymine leads to slower condensation. Consequently, substantial quantities of the unwanted α-linked nucleoside resulted. The desired β-isomer is difficult to separate from the mixture; in Chattopadhyaya's synthesis the protecting groups were removed before separation of the isomers by ion exchange. We chose an indirect route involving initial preparation of protected deoxyuridines **12a–b** which were obtained in high yield (84–92%) in a manner analogous to the thymidine synthesis. Several methods were then explored for conversion of the deoxyuridine to deoxycytidine (Scheme III). In the preparation of 1′-deuteriodeoxycytidine (**14a**), protected 1′-d_1-deoxyuridine **12a** was converted to 4-chloro derivative **13** by treatment with thionyl chloride/DMF (61% yield plus 25% recovered starting material), a procedure first used by Zemlicka and Sorm[28] for the synthesis of some related chlorouridine derivatives. Ammonolysis gave deoxycytidine **14a** in 95% yield. The chlorination reaction required careful monitoring and had to be quenched while considerable starting material remained to avoid excessive degradation; therefore a route based on triazolide chemistry[29] was explored as an alternative. Deoxyuridine derivative **12a** was converted to triazolide **15** by treatment with triazole and $POCl_3$; **15** was treated with NH_4OH in dioxane followed by NH_3/MeOH to give **14a** in 44% overall yield from **12a**. In the synthesis of 1′,2′,2″-trideuteriodeoxycytidine (**14b**), deoxyuridine derivative **12b** was converted to the 4-(3-nitro)-1,2,4-triazolide derivative (**17**) (67% yield + 13% recovered starting material). Ammonolysis of **17** gave **14b** in 67% yield. Some of the 4-*O*-methyl derivative was formed during ammonolysis, a problem which can be avoided by the two-step procedure used for conversion of **15** to **14a**. The best route from **12** to **14** in terms of yield appears to be through the 4-chloro derivative, but the three-step triazolide procedure is the simplest in terms of execution and purification. It is likely that none of these procedures has been fully optimized and further improvements are possible. The synthesis by Chattapadhyaya[12] gave deoxycytidine in 40% yield.

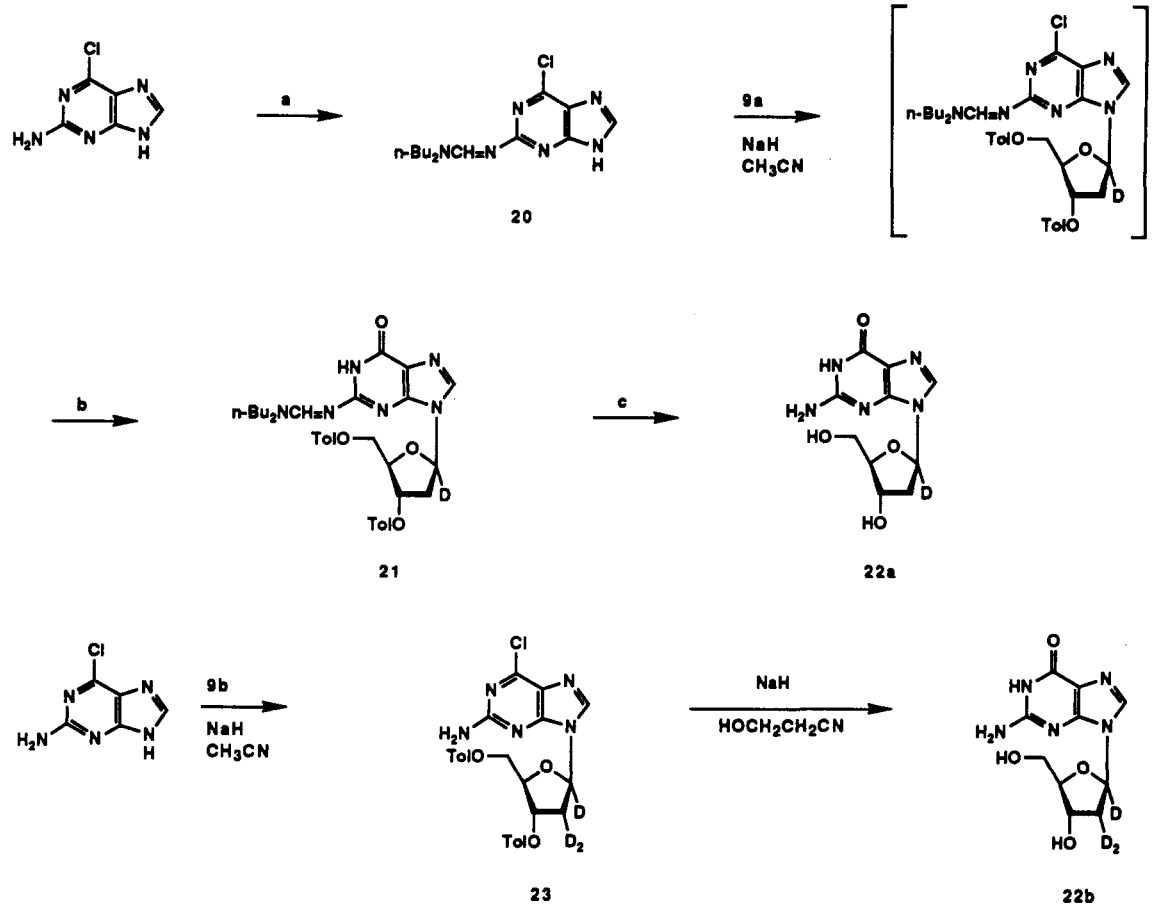
(27) Hubbard, A. J.; Jones, A. S.; Walker, R. T. *Nucleic Acids Res.* **1984**, *12*, 6827.
(28) Zemlicka, J.; Sorm, F. *Collect. Czech. Chem. Commun.* **1965**, *30*, 2052.
(29) (a) Divakar, K. J.; Reese, C. B. *J. Chem. Soc., Perkin Trans. 1* **1982**, 1171. (b) Reese, C. B.; Ubasawa, A. *Tetrahedron Lett.* **1980**, *21*, 2265. (c) Kraszewski, A.; Stawinski, J. *Tetrahedron Lett.* **1980**, *21*, 2935.

Scheme IV[a]



[a] (a) NaH, CH_3CN; (b) NH_3, MeOH.

Scheme V[a]



[a] (a) n-$Bu_2NCH(OEt)_2$; (b) NaH, pyridine-2-carbaldoxime; (c) NH_3, MeOH.

The purine deoxynucleosides were synthesized by methodology developed by Robins[30] in which sodium salts of halopurines are condensed with Hoffer's α-chloro sugar derivative; 6-chloropurine and 2-amino-6-chloropurine were used for synthesis of deoxyadenosine and deoxyguanosine, respectively. Generally, only traces of the 9α anomers are formed, and the main impurity, the 7β regioisomer, is easily removed by chromatography.

To prepare deuteriated 2′-deoxyadenosine, the sodium salt of 6-chloropurine was treated with α-chloro sugar (**9a** or **9b**) in CH_3CN to give purine derivatives **18a** and **18b** which upon treatment with methanolic ammonia gave deuteriated deoxyadenosines **19a** and **19b** in 37–45% overall yield for the two step procedure (Scheme IV).

The synthesis of deoxyguanosine proved to be more troublesome. A study of literature examples revealed a number of variants on the basic strategy, none of which gave satisfactory yields.[31] Pathak et al.[12a] and Wu et al.[12b] obtained a 51% yield of the 9β adduct by reaction of the sodium salt of 2-amino-6-chloropurine with the α-chloro sugar. Hydrolytic removal of the chloro substituent presented problems for purification of the product and,

(30) (a) Kazimierczuk, Z.; Revankar, G. R.; Robins, R. K. *Nucleic Acids Res.* **1984**, *12*, 1179. (b) Kazimierczuk, Z.; Cottam, H. B.; Revankar, G. R.; Robins, R. K. *J. Am. Chem. Soc.* **1984**, *106*, 6379. (c) Revankar, G. R.; Gupta, P. K.; Adams, A. D.; Dalley, N. K.; McKernan, P. A.; Cook, D. P.; Canonico, P. G.; Robins, R. K. *J. Med. Chem.* **1984**, *27*, 1389.

(31) (a) Iwamoto, R. H.; Acton, E. M.; Goodman, L. *J. Med. Chem.* **1963**, *6*, 684. (b) Lee, W. W.; Martinez, A. P.; Goodman, L.; Henry, D. W. *J. Org. Chem.* **1972**, *37*, 2923. (c) Robins, M. J.; Robins, R. K. *J. Org. Chem.* **1969**, *34*, 2160. (d) Robins, M. J.; Khwaja, T. A.; Robins, R. K. *J. Org. Chem.* **1970**, *35*, 636. (e) Mengel, R.; Muhs, W. *Chem. Ber.* **1979**, *112*, 625. (f) Venner, H. *Chem. Ber.* **1960**, *93*, 140.

after treatment with either trimethylamine–tetraethylammonium hydroxide or sodium methoxide/2-mercaptoethanol, they chose to isolate deoxyguanosine as the triacetate rather than as the free nucleoside.

For the synthesis of 1'-deuteriodeoxyguanosine we utilized a different strategy (Scheme V). 2-Amino-6-chloropurine was protected at N-2 with the *N,N*-di-*n*-butylformamidino group[32] to increase solubility of the purine and to yield a glycosylation product from which chloride might be more easily displaced by oxygen nucleophiles. The group would also block potential side reactions at the 2-position. The sodium salt of formamidino derivative **20**, prepared with sodium hydride in acetonitrile, was treated with α-chloro sugar **9a**. After 10 min, the reaction was assumed to be complete and the sodium salt of pyridine-2-carbaldoxime was added to displace the chloro group from C-6; pyridine-2-carbaldoxime is commonly used in nucleoside chemistry for cleavage of *p*-chlorophenyl groups from phosphotriester oligonucleotides.[33] Workup by chromatography gave the desired 9β adduct, **21**, in 29% yield. Other major products included the 7β isomer and the 3β isomer formed in the ratio of 1.0:0.9:0.5 of 9β:7β:3β. Ammonolysis of compound **21** gave 1'-deuteriodeoxyguanosine (**22a**) in 91% yield.

The relatively low yield of the desired 9β product in this reaction led us to repeat Chattopadhyaya's direct coupling of 2-amino-6-chloropurine by Robins' procedure. This reaction, when carried out with trideuteriated chloro sugar **9b** gave the 9β adduct **23** in 54–61% purified yield. The 7β isomer is also formed in this reaction (9β:7β, ~3:1) but is easily separated by chromatography. Displacement of the 6-chloro substituent was initially attempted with pyridine-2-carbaldoxime; however, a complex mixture of products was obtained, perhaps due to ionization of the 2-amino group. An alternative route was examined involving formation of the 6-methoxy derivative using sodium methoxide,[34] followed by cleavage of the methyl ether with trimethylsilyl iodide, a procedure used in very high yield by Ramaswamy et al. for the preparation of 7-deazadeoxyguanosine.[35] The methoxide displacement proceeded smoothly in 98% yield, but extensive depurination occurred during demethylation with trimethylsilyl iodide, even in the presence of acid scavengers.

Although conditions could not be found for clean demethylation of the 6-methoxy derivative, the high yield obtained in the displacement reaction encouraged us to attempt preparation of other 6-alkoxy derivatives which might be dealkylated more readily. The 2-cyanoethoxy group, widely used as a transient protecting group in nucleotide chemistry,[36] was chosen because it is easily removed by β-elimination with ammonium hydroxide. When **23** was treated with excess sodium 2-cyanoethoxide, the only detectable nucleoside product was deuteriated deoxyguanosine (**22b**); sodium 2-cyanoethoxide caused detoluoylation in addition to in situ β-elimination of the 6-(2-cyanoethyl) ether. The deoxyguanosine was isolated in 50–60% yield by ion-exchange chromatography. This procedure for the synthesis of 2'-deoxyguanosine, although not as high yielding as those for the other nucleosides, represents a substantial improvement over previously reported methods.

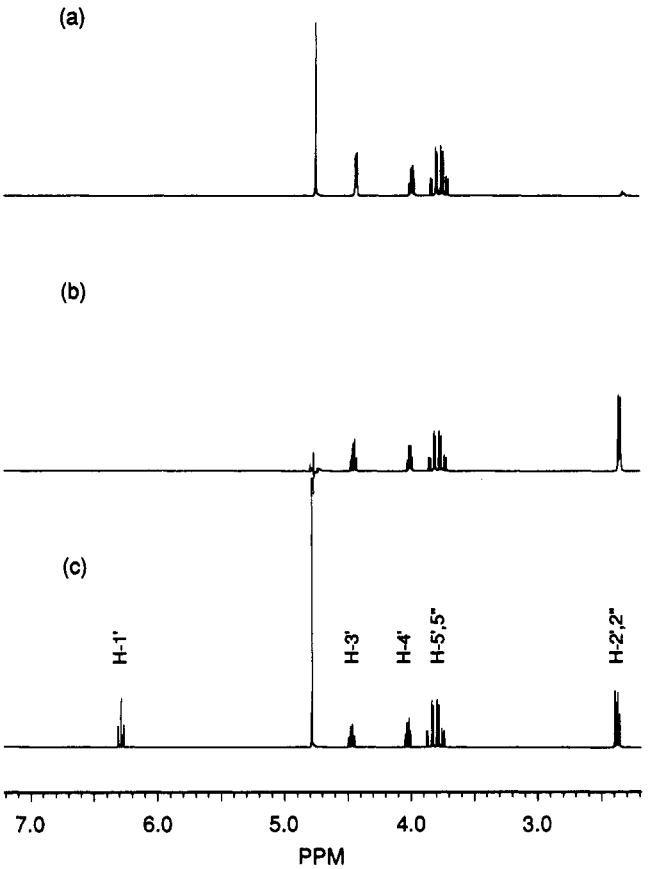


Figure 1. ^{1}H NMR spectra of the deoxyribose region of thymidine: (a) 1',2',2"-trideuteriothymidine, (b) 1'-deuteriothymidine, (c) undeuteriated thymidine.

Figure 1 shows the ^{1}H NMR spectra of 2'-deoxythymidine, and its 1'-deuterio and 1',2',2"-trideuterio derivatives as examples of the simplification in ^{1}H spectra introduced by deuteriation. Not only is the deuteriated signal removed from the spectrum but also the remaining spectrum is simplified by elimination of ^{1}H–^{1}H spin coupling from the vicinal positions.

In summary, we have developed efficient routes for introducing deuterium into the 1- and 2-positions of deoxyribose and have used the deuteriated deoxyribose to prepare deoxyribonucleosides. Improved procedures for the preparation of 2'-deoxycytidine and 2'-deoxyguanosine have been developed. The preparation of specifically deuteriated oligodeoxynucleotides is now underway to demonstrate the utility of deuteriation in simplification of two-dimensional NMR spectra.

Experimental Section

NMR spectra were recorded on Bruker AM-400 and AF-300 spectrometers: ^{1}H at 300 or 400, ^{13}C at 100, and ^{2}H at 61 MHz. For ^{1}H spectra tetramethylsilane was used as internal standard in organic solvents; sodium 2,2-dimethyl-2-silapentane-5-sulfonate was used in D_2O. ^{2}H NMR spectra were acquired in H_2O with HDO (4.63 ppm) as internal standard or in freshly distilled chloroform with $CDCl_3$ (7.27 ppm) as internal standard. Mass spectra were obtained on Nermag 1010C and VG 70-250 HF spectrometers. Fast atom bombardment (FAB) spectra were acquired on the latter instrument with 3-nitrobenzyl alcohol as the matrix solvent. Melting points are uncorrected. TLC spots

(32) (a) Froehler, B. C.; Matteuci, M. D. *Nucleic Acids Res.* **1983**, *11*, 8031. (b) McBride, L. J.; Kierzek, R.; Beaucage, S. L.; Caruthers, M. H. *J. Am. Chem. Soc.* **1986**, *108*, 2040.

(33) (a) Robins, M. J.; Robins, R. K. *J. Am. Chem. Soc.* **1965**, *87*, 4934. (b) Reese, C. B.; Zard, L. *Nucl. Acids Res.* **1981**, *9*, 4611.

(34) (a) Milne, G. H.; Townsend, L. B. *J. Med. Chem.* **1974**, *17*, 263. (b) Mehta, J. R.; Ludlum, D. B. *Biochim. Biophys. Acta* **1978**, *521*, 770. (c) Gaffney, B. L.; Jones, R. A. *Tetrahedron Lett.* **1982**, *23*, 2253. (d) Bernadou, J.; Blandin, M.; Meunier, B. *Nucleosides Nucleotides* **1983**, *2*, 459.

(35) Ramasamy, K.; Imamura, N.; Robins, R. K.; Revankar, G. R. *Tetrahedron Lett.* **1987**, *28*, 5107.

(36) (a) Schaller, H.; Weimann, G.; Lerch, B.; Khorana, H. G. *J. Am. Chem. Soc.* **1963**, *85*, 3821. (b) Sinha, N. D.; Biernat, J.; McManus, J. Köster, H. *Nucleic Acids Res.* **1984**, *12*, 4539. (c) Gaffney, B. L.; Jones, R. A. *Tetrahedron Lett.* **1982**, *23*, 2257.

were routinely detected by UV and a *p*-anisaldehyde spray reagent.[37] Elemental analyses were performed by Galbraith Laboratories, Knoxville, TN.

Tetrahydrofuran (THF) was purified by distillation from a sodium–potassium alloy containing benzophenone ketyl. Other solvents were distilled from the indicated drying agent: CH_3CN (CaH_2), MeOH (Mg turnings), Et_2O ($LiAlH_4$), $CHCl_3$ (P_2O_5), CH_2Cl_2 (CaH_2), and pyridine (KOH). Organic extracts were routinely dried over $MgSO_4$ before evaporation.

2,2′-Dideuterio-2-deoxyribonolactone (3b). Methanol-*d* was prepared by hydrolysis of trimethyl orthoformate or tetramethyl orthosilicate with D_2O in the presence of a trace of *p*-toluenesulfonic acid using a 5% excess of the ortho ester. The MeOD was collected by distillation and carefully fractionally distilled, bp 64–66 °C. Tetramethyl orthosilicate was found to be the more satisfactory source of MeOD because traces of methyl formate contaminated material prepared from methyl orthoformate.

2-Deoxyribonolactone (**3a**)[16,17] (17.5 g, 0.132 mol) [^{1}H NMR (d_4-MeOH) δ 5.90 (br s, OH's, 2), 4.46 (dt, H3, 1), 4.40 (q, H4, 1), 3.80 (dd, H5, 1), 3.71 (dd, H5′, 1), 2.95 (dd, H2, 1), 2.40 (dd, H2′, 1); $J_{2,2'}$ = 18.0 Hz, $J_{2,3}$ = 6.8, $J_{2',3}$ = 2.6, $J_{3,4}$ = 2.3, $J_{4,5}$ = 3.3, $J_{4,5'}$ = 3.5, $J_{5,5'}$ = 12.4 Hz; ^{13}C NMR (D_2O; CH_3CN as reference = 1.3 ppm) δ 179.7, 89.2, 68.5, 61.3, 38.0] was lyophilized twice from 40 mL of D_2O (99% D) to exchange the hydroxyl protons. This sample was coevaporated twice with dry pyridine, dried in vacuo, and then dissolved in 130 mL of freshly prepared MeOD. The reaction was allowed to stand for 7 days at ambient temperature; aliquots of NaOMe/MeOD were added on a daily basis; total amount: 13.9 mmol (0.105 equiv). The reaction was quenched by evaporation of the solvent and treatment at 0 °C with CF_3CO_2D (65 mL). The solution was heated for 1 h at 70 °C, evaporated under reduced pressure, coevaporated twice with pyridine, and finally dried in vacuo to give 15.08 g (85%) of **3b** as a clear oil. Deuteriation: 95% D at H-2′ and 87% at H-2 as monitored by ^{1}H NMR. ^{1}H NMR (400 MHz, D_2O) was essentially unchanged except for reduction of areas of the signals for H2 and H2′ to 0.13 and 0.05, respectively, and simplification of the signal for H3 to a doublet. ^{13}C NMR (100 MHz, D_2O) unchanged except that the signal at 38.0 had become very weak and showed splitting. ^{2}H NMR (H_2O): δ 2.82 (br s, D2′), 2.35 (br s, D2). MS (FAB$^+$): *m/z* 157 (M + Na), 135 (M + H), 117 (MH – H_2O).

Undeuteriated and 2,2′-Dideuterio-3,5-bis-*O*-(*tert*-butyldimethylsilyl)-2-deoxyribonolactones (4a and 4b). *tert*-Butyldimethylsilyl nitrate (0.223 mmol) was prepared from the chloride and $AgNO_3$ in 200 mL of THF under argon.[19] Dry pyridine (54.1 mL, 0.669 mol) was added, and after 10 min, 13.4 g (0.101 mol) of **3a** was added in 150 mL of dry THF. The mixture was stirred for 8 h and then filtered; the filter cake was washed with CH_2Cl_2. The filtrate and washings were combined, evaporated, and diluted in Et_2O. Precipitated material was filtered off, and the residue was washed with Et_2O. The combined filtrates were washed twice with H_2O, dried, and evaporated in vacuo to yield 36.5 g (99%) of **4a** as a white crystalline powder, mp 78–80 °C and 80–81 °C after sublimation (95 °C, 0.25 mmHg). ^{1}H NMR ($CDCl_3$): δ 4.48 (dt, H3, 1), 4.30 (q, H4, 1), 3.79 (dd, H5, 1), 3.73 (dd, H5′, 1), 2.80 (dd, H2, 1), 2.35 (dd, H2′, 1), 0.87 (s, C-Me, 18), 0.06 (s, Si-Me, 6), 0.05 (s, Si-Me, 3), 0.04 (s, Si-Me, 3); $J_{2,2'}$ = 17.7, $J_{2,3}$ = 2.5, $J_{2',3}$ = 2.5, $J_{3,4}$ = ~2.5, $J_{4,5}$ = 3.2, $J_{4,5'}$ = 2.5, $J_{5,5'}$ = 11.5 Hz. ^{13}C NMR ($CDCl_3$): δ 175.6, 88.0, 69.6, 62.4, 38.9, 25.8, 25.6, 18.2, 17.9, -4.8, -4.9, -5.6, -5.7. MS (FAB$^+$): *m/z* 361 (M + H), 303 (M – C_4H_9). HRMS (EI): calcd for $C_{13}H_{27}O_4Si_2$ (M – C_4H_9) 303.1448, found 303.1460. $[\alpha]^{21}_D$ = +12.1°. Anal. Calcd for $C_{13}H_{27}O_4Si_2$: C, 56.62; H, 10.06. Found: C, 56.36; H, 10.06.

The 2,2′-d_2 analogue **4b** was prepared by the same procedure in 88% yield. The ^{1}H NMR spectrum showed reduction in the area of the signals at 2.80 and 2.35 to 0.06 and 0.14, respectively. The signal at 4.48 underwent simplification. The ^{13}C spectrum was identical with that of **4a** except the signal at 38.9 (CH_2) was weak; in addition, new signals were present at 38.7 (CHD) and 38.4 (CD_2). ^{2}H NMR showed signals for the D-2 and D-2′. MS (FAB$^+$): *m/z* 363 and 305. HRMS (EI): calcd for $C_{13}H_{25}D_2O_4Si_2$ (M – C_4H_9) 305.1571, found 305.1564.

1-Deuterio- (5a) and 1,2,2′-Trideuterio-3,5-di-*O*-(*tert*-butyldimethylsilyl)-2-deoxyribose (5b). 2-Deoxyribonolactone (**4a**, 5.0 g, 13.86 mmol) in 250 mL of CH_2Cl_2 was treated with 10.4 mL of a 1.4 M solution of diisobutylaluminum deuteride in hexane–Et_2O (14.56 mmol, 1.05 equiv, prepared by the method of Kalvin and Woodard and titrated just before use by reduction of benzophenone monitored by TLC)[20b] for 2 h at -78 °C under argon. The reaction was monitored by TLC (2% MeOH/CH_2Cl_2) until disappearance of all starting material or formation of diol **6a** occurred. The reaction was quenched with MeOH (10 mL). The mixture was allowed to come to room temperature by stirring overnight. Workup involved filtration and washing the filtrate twice with sodium potassium tartrate solution. Emulsions were broken by filtering through Celite. The organic solution was dried and evaporated in vacuo. The residue was partitioned by flash chromatography on silica gel (CH_2Cl_2–MeOH gradient up to 1% MeOH) to give 4.18 g (83%) of **5a** as a clear oil. ^{1}H NMR showed a 2:1 mixture of α and β anomers. ^{1}H NMR ($CDCl_3$) α anomer: δ 4.34 (d, H3, 1), 4.15 (dd, H4, 1), 3.96 (s, OH, 1), 3.58 (dd, H5, 1), 3.28 (dd, H5′, 1), 2.00 (dd, H2, 1), 1.88 (dd, H2′, 1), 0.85 (s, C-Me, 9), 0.84 (s, C-Me, 9), 0.07 (s, Si-Me, 3), 0.01 (s, Si-Me, 3), 0.00 (s, Si-Me, 3), -0.01 (s, Si-Me, 3); $J_{2,2'}$ = 13.3, $J_{2',3}$ = 0.7, $J_{2,3}$ = 4.7, $J_{3,4}$ = ~0, $J_{4,5}$ = 4.0, $J_{4,5'}$ = 7.2, $J_{5,5'}$ = 10.7 Hz. β anomer: δ 4.45 (ddd, H3, 1), 3.91 (q, H4, 1), 3.77 (s, OH, 1), 3.68 (dd, H5, 1), 3.64 (dd, H5′, 1), 2.08 (dd, H2, 1), 1.99 (dd, H2′, 1), 0.87 (s, C-Me, 9), 0.83 (s, C-Me, 9), 0.07 (s, Si-Me, 3), 0.06 (2 s, Si-Me, 6), 0.03 (s, Si-Me, 3); $J_{2,2'}$ = 13.3, $J_{2',3}$ = 5.7, $J_{2,3}$ = 6.8, $J_{3,4}$ = 3.5, $J_{4,5}$ = 3.5, $J_{4,5'}$ = 3.1, $J_{5,5'}$ = 10.7 Hz. ^{13}C NMR ($CDCl_3$) δ 87.3 (α), 87.2 (β), 74.1 (α), 72.2 (β), 63.7 (β), 63.4 (β), 44.9 (β), 41.2 (α), 25.9 (α + β), 25.7 (α + β), 18.3 (α), 17.9 (β), -4.8 (α or β), -5.0 (α + β), -5.5 (2) (α + β). ^{2}H NMR (64.1 MHz, $CHCl_3$) δ 5.37 (br s, D1, α + β); MS (FAB$^+$) *m/z* 386 (M + Na$^+$), 346 (M – OH), 306, 214, 133, 115. HRMS (EI): calcd for $C_{17}H_{36}DO_3Si_2$ (M – OH) 346.2342, found 346.2342; calcd for $C_{13}H_{28}DO_4Si_2$ (M – C_4H_9) 306.1666, found 306.1664.

Compound **5b**, prepared (82% yield) by the procedure used for **5a**, was a clear oil. The ^{1}H NMR was identical with that of **5a** except that the signals for the protons on C2 were diminished to ca. 10% of their former intensity and broadened by geminal deuterium coupling; the signals for H3 showed reduced multiplicity: α anomer δ 4.38 (~s), β anomer 4.49 (d, $J_{3,4}$ = 3.7). In the ^{13}C NMR spectrum the signals for the C2 at 44.9 (β) and 41.2 (α) appeared as weak multiplets. ^{2}H NMR ($CHCl_3$): δ 5.35 (br s, D1, α + β), 2.00 (br s, D2, α + β and D2′, β), 1.89 (br s, D2′, α). MS (FAB$^+$): *m/z* 348 (M – OH), 215, 133, 115. HRMS (EI): calcd for $C_{17}H_{34}D_3O_3Si_2$ (M – OH) 348.2468, found 348.2476; calcd for $C_{13}H_{26}D_3O_4Si_2$ (M – C_4H_9) 308.1789, found 308.1800.

Methyl 1-Deuterio- and 1,2,2′-Trideuterio-2-deoxyriboside (7a–b). Compound **5a** (5.0 g, 13.75 mmol) was stirred in 100 mL of 0.1% methanolic HCl for 15 min at ambient temperature.[38] Powdered Ag_2CO_3 (1.5 g) was added; the mixture was filtered to remove salts and evaporated in vacuo to leave 3.26 g of residue. ^{1}H NMR indicated complete conversion to methyl glycoside. The product was largely in the furanose form. Partial loss of silyl groups had occurred (39% remaining silyl groups). The material was treated with 0.2 M tetra-*n*-butylammonium fluoride in THF (180 mL) at ambient temperature for 30 min. TLC (5% MeOH/CH_2Cl_2) indicated complete removal of silyl groups. The solution was passed through a short column of silica gel which was washed with 10% MeOH/CH_2Cl_2. The effluent was concentrated and partitioned on a C_{18} reverse-phase column using H_2O–MeOH mixtures (up to 30% MeOH) to give **7a** as a clear oil (1.85 g, 90%). ^{1}H NMR ($CDCl_3$): ~1:1 mixture of anomers δ 4.48 (m, 1), 4.18 (m, 1), 4.09 (q, 1), 4.03 (q, 1), 3.66 (m, 4), 3.39 (s, OCH_3, 3), 3.36 (s, OCH_3, 3), 2.18 (m, 3), 1.97 (dd, 1, *J* = 11.9, 2.0 Hz). ^{13}C NMR ($CDCl_3$): δ 104.2 (C2, weak multiplet), 86.6, 85.7, 71.1, 70.8, 62.0, 61.7, 53.8, 53.5, 40.9, 40.4. ^{2}H NMR ($CHCl_3$): δ 5.11 (br s, D1, α + β). MS (FAB$^+$): *m/z* 172 (M + Na), 118, 100.

Compound **5b** was converted to **7b** (clear oil, 3.23 g, 97%). The ^{1}H NMR was similar to **7a** except that the signals at 2.18 and 1.91 were of reduced intensity (~0.10). In the ^{13}C spectrum the C2 signals at 40.9 and 40.4 were weak multiplets. ^{2}H NMR (64.1

(37) *Thin Layer Chromatography*; Stahl, E., Ed.; Springer-Verlag: New York, 1969; p 857.

(38) Deriaz, R. E.; Overend, W. G.; Stacey, M.; Wiggins, L. F. *J. Chem. Soc.* **1949**, 2836.

MHz, $CHCl_3$): δ 5.10 (br s, D1, α + β, 2), 2.11 (br s, D2, α + β, 2), 1.98 (br s, D2′, β, 1), 1.85 (br s, D2, α, 1). MS (FAB⁺): *m/z* 174 (M + Na), 120, 101.

Methyl 1-Deuterio- and 1,2,2′-Trideuterio-3,5-di-*O*-*p*-toluoyl-2-deoxyriboside (8a–b). Compound **7a** (1.70 g, 11.37 mmol) in 50 mL of pyridine containing 4-(dimethylamino)pyridine (5 mg) was treated with 3.16 mL of *p*-toluoyl chloride (23.8 mmol) for 2 h at 40 °C. The reaction was quenched by slow addition of 10 mL of H_2O at 0 °C. An Et_2O extract was washed with brine, dried, and evaporated in vacuo. Recrystallization from MeOH gave 3.6 g of **8a**. The mother liquor was further processed by flash chromatography on silica gel eluted with 1% MeOH/CH_2Cl_2 to give 0.59 g of additional **8a**. Total yield: 4.19 g (95%). Mp: 66–68 °C (lit.[23] mp 76.5 °C). ^1H NMR ($CDCl_3$): anomeric mixture δ 7.95 (m, 8), 7.23 (m, 8), 5.61 (m, 1), 5.42 (m, 1), 4.67–4.45 (m, 6), 3.44 (s, 3), 3.37 (s, 3), 2.59–2.52 (m, 2), 2.40 (4 overlapping s, 12), 2.34 (dd, 1, *J* = 5.1, 14.1 Hz), 2.19 (dd, 1, *J* = 2.1, 14.6 Hz). ^{13}C NMR ($CDCl_3$): δ 166.5, 166.3, 166.1, 144.0, 143.9, 143.7, 143.6, 130.0, 129.8, 129.7, 129.1, 127.3, 127.1, 126.9, 105.0 (weak multiplet), 81.9, 80.9, 75.4, 74.6, 65.1, 64.3, 55.1, 55.0, 39.2, 21.6. ^2H NMR ($CHCl_3$): δ 5.20 (br s, D1, α + β). MS (FAB⁺): *m/z* 408 (M + Na), 354, 218, 119, 82.

Compound **7b** was converted to **8b** by the same procedure in 95% yield, mp 66–68 °C. ^1H NMR ($CDCl_3$): δ (α/β ratio ~ 1.0/1.3) 8.00–7.90 (m, aromatic, 8), 7.26–7.20 (m, aromatic, 8), 5.60 (d, $H3_\alpha$, 1, *J* = 2.4 Hz), 5.40 (d, $H3_\beta$, 1, *J* = 3.5 Hz), 4.66–4.44 (m, $H4,5,5'_{\alpha+\beta}$, 6), 3.44 (s, $OCH_{3\beta}$, 3), 3.38 (s, $OCH_{3\alpha}$, 3), 2.42 (2 overlapping singlets, toluoyl $CH_{3\,\alpha+\beta}$, 12). ^{13}C NMR ($CDCl_3$): δ 166.5, 166.3, 166.1, 144.0, 143.9, 143.7, 143.6, 130.2, 129.8, 129.7, 129.1, 127.3, 127.1, 127.0, 81.9, 80.9, 75.4, 74.5, 65.1, 64.3, 55.1, 55.0, 38.6 (C2 weak multiplet), 21.6. ^2H NMR ($CHCl_3$): δ 5.19 (br s, $D1_{\alpha+\beta}$, 2), 2.52 (br s, $D2_{\alpha+\beta}$, 2), 2.31 (br s, $D2_\beta$, 1), 2.16 (br s, $D2_\alpha$, 1). MS (FAB⁺): *m/z* 410 (M + Na), 356, 219, 119, 83. Anal. Calcd for $C_{22}H_{21}D_3O_6$: C, 68.16; H/D, 6.24. Found: C, 68.25; H/D, 6.14.

1-Deuterio- (9a) and 1,2,2′-Trideuterio-3,5-di-*O*-*p*-toluoyl-2-deoxyribosyl Chloride (9b). Compound **9a–b** (lit.[23] mp 109 °C) was prepared from **8a–b** in 66% yield by the procedure of Hoffer.[23] The melting points of various batches of α-chloro sugar varied between 110 and 130 °C, but the melting range was always narrow and no correlation of melting point with subsequent condensation yields was seen as long as the material was kept stored over drying agent at −20 °C and used as promptly as possible. Anomerization is catalyzed by HCl from decomposition. Old chloro sugar can be reconverted to the starting methyl glycoside by treatment with MeOH. **9a**: ^1H NMR ($CDCl_3$): δ 7.95 (m, aromatic, 4), 7.24 (m, aromatic, 4), 5.56 (m, H3, 1), 4.86 (m, H4, 1), 4.69 (dd, H5, 1), 4.59 (dd, H5′, 1), 2.87 (dd, H2, 1), 2.74 (d, H2′, 1), 2.43 (s, CH_3, 3), 2.41 (s, CH_3, 3); $J_{2,2'}$ = 14.9, $J_{2,3}$ = 7.3, $J_{2',3}$ = ~0, $J_{3,4}$ not determined, $J_{4,5}$ = 3.2, $J_{4,5'}$ = 4.3, $J_{5,5'}$ = 12.1 Hz. ^{13}C NMR ($CDCl_3$): δ 166.31, 165.98, 144.20, 143.96, 129.85, 129.62, 129.16, 126.79, 126.67, 95.00 (weak t, C1′), 84.62, 63.44, 44.34, 21.61. ^2H NMR (CH_2Cl_2): δ 6.42 (D1′). MS (FAB⁺): *m/z* 354 (M − Cl), 119.

9b was prepared as above (83% yield), mp 120–121 °C dec. The ^1H NMR differed from that of **9a** in that only residual signals were observed for H2 and H2′; H3 appeared as a doublet ($J_{3,4}$ = 2.7 Hz). The ^{13}C spectrum was missing the signal at 44.34. ^2H NMR ($CHCl_3$): δ 6.44 (br s, D1), 2.71 (m, D2 and D2′). MS (FAB⁺): *m/z* 356 (M − Cl), 119. Anal. Calcd for $C_{26}H_{20}D_3N_4O_5Cl$: C, 60.26; H/D, 4.55; N, 10.99. Found: C, 60.24; H/D, 4.50; N, 10.94.

1′-Deuterio- (10a) and 1′,2′,2″-Trideuterio-3′,5′-di-*O*-*p*-toluoylthymidine (10b). The procedure of Hubbard et al.[27] was followed for the synthesis of pyrimidine nucleosides. Thymine (0.195 g, 1.54 mmol) was refluxed overnight in a mixture of chlorotrimethylsilane (3 mL) and hexamethyldisilazane (7 mL). The solution was allowed to cool, and the volatile materials were removed by distillation under aspirator vacuum. The residue was dissolved in 20 mL of $CHCl_3$, which had been freshly distilled from P_2O_5. Chloro sugar **9a** (0.3 g, 0.77 mmol) was added as a solid all at once, and the reaction mixture was allowed to stir for 2 h. MeOH (0.1 mL) was added, and the excess thymine which precipitated was removed by filtration. The solvent was evaporated, and the residue was recrystallized from EtOAc. The mother liquor was evaporated and the residue purified by preparative TLC (CH_2Cl_2–MeOH, 92:8). The band corresponding to the product was scraped from the plate, eluted with CH_2Cl_2–MeOH, (80:20), and combined with the recrystallized material to give a total yield of 0.37 g (91%). A sample of **10a** was recrystallized from EtOAc, mp 196–197.5 °C (lit.[23b] mp 197 °C). ^1H NMR ($CDCl_3$): δ 8.40 (br s, H3, 1), 7.94 (m, aromatic, 4), 7.28 (m, aromatic and H6, 5), 5.64 (~dt, H3′, 1), 4.78, (dd, H5′, 1), 4.65 (dd, H5″, 1), 4.52 (m, H4′, 1), 2.69 (dd, H2″, 1), 2.44 (s, CH_3, 3), 2.43 (s, CH_3, 3), 2.31 (dd, H2′, 1), 1.62 (s, 5-CH_3, 3); $J_{2',2''}$ = 14.3, $J_{2',3'}$ = 6.6, $J_{2'',3'}$ = 1.5, $J_{3',4'}$ = ~1.5, $J_{4',5'}$ = 3.0, $J_{4',5''}$ = 3.0, $J_{5',5''}$ = 12.3 Hz. ^{13}C NMR ($CDCl_3$): δ 166.01, 163.41, 150.26, 144.55, 134.40, 129.83, 129.51, 129.46, 129.28, 126.58, 126.30, 111.65, 82.79, 74.88, 64.16, 37.91, 21.69, 12.09. ^2H NMR (CH_2Cl_2): δ 6.39 (br s, D1′). MS (CI⁺): *m/z* 480 (M + H) 354, 137, 127, 119. UV (CH_3CN): λ_{max} (ε) 240 (36 200), 266 (sh) nm.

Compound **10b** was prepared as described above for **10a** to give 640 mg, mp 198–202 °C, in the first crop. Evaporation of the mother liquor and recrystallization from EtOAc gave an additional 120 mg of **10b**, mp 195–198 °C. Total yield of **10b** was 81%. The NMR spectrum was identical with that of **10a** except that only residual signals were observed for H2′ and H2″; the signal for H3′ was a doublet ($J_{3',4'}$ = 1.9 Hz). The ^{13}C NMR spectrum was missing the signal at 37.91. ^2H NMR ($CHCl_3$) δ 6.45 (br s, D1′), 2.67 (br s, D2″), 2.30 (br s, D2′). MS (FAB⁺): *m/z* 482 (M + H), 356, 119.

1′-Deuterio- (11a) and 1′,2′,2″-Trideuteriothymidine (11b). Compound **10a** (0.5 g, 1.04 mmol) was treated for 2 days at room temperature with MeOH (30 mL) which had been saturated with NH_3 gas at 0 °C. The solvent was evaporated; the residue was dissolved in H_2O and washed with Et_2O. The H_2O was evaporated, and the residue was dried in vacuo. Trituration with EtOAc/Et_2O gave a white solid (0.244 g, 97%). Recrystallization from H_2O gave colorless crystals of **11a**, mp 185–186.5 °C (lit.[39] mp 182–183 °C). ^1H NMR (D_2O): δ 7.66 (m, H6, 1), 4.48 (m, H3′, 1), 4.03 (m, H4′, 1), 3.85 (dd, H5′, 1), 3.78 (dd, H5″, 1), 2.39 (dd, H2″, 1), 2.35 (dd, H2′, 1), 1.89 (d, CH_3, 3); $J_{2',2''}$ = 12.4, $J_{2',3'}$ = 7.1, $J_{2'',3'}$ = 4.6, $J_{3',4'}$ not determined, $J_{4',5'}$ = 3.6, $J_{4',5''}$ = 5.0, $J_{5',5''}$ = 12.4; $J_{5\text{-Me},6}$ = 1.2 Hz. ^{13}C NMR (D_2O): δ 167.10, 152.35, 138.22, 112.12, 87.31, 85.57 (t, C1′, $^1J_{C,D}$ = 26 Hz), 71.25, 62.00, 39.23, 12.31. ^2H NMR (H_2O): δ 6.05 (br s, D1′). MS (CI⁺): *m/z* 244 (M + H), 127, 118, 100, 82. MS (FAB⁺): *m/z* 336 (M+H+glycerol), 244 (M+H), 127, 118. UV (H_2O): λ_{max} (ε) 266 nm (9450).

Ammonolysis of **10b** (700 mg, 1.45 mmol) gave 335 mg (99%) of **11b**. The ^1H NMR spectrum had only residual signals for H2′ and H2″; H3′ gave a doublet ($J_{3',4'}$ = 3.9 Hz). The ^{13}C NMR spectrum had a weak multiplet at 38.5 replacing the signal at 39.23 in **10a**. ^2H NMR (H_2O): δ ppm 6.06 (br s, D1′), 2.14 (br s, D2′ and D2″). MS (FAB⁺): *m/z* 246 (M + H), 127, 120.

1′-Deuterio- (12a) and 1′,2′,2″-Trideuterio-3′,5′-di-*O*-*p*-toluoyluridine (12b). Uracil (0.576 g, 5.14 mmol) was refluxed in a mixture of 3 mL of chlorotrimethylsilane and 7 mL of hexamethyldisilazane for 4 h. Another 10 mL of the silanizing mixture was added, and refluxing was continued for 4 more h to give 2,4-bis((trimethylsilyl)oxy)pyrimidine. Volatile materials were removed by distillation. The residue was dissolved in 30 mL of $CHCl_3$. Chloro sugar **9a** (0.9 g, 2.31 mmol) was added as a solid all at once and the mixture was stirred for 2 h. MeOH (0.1 mL) was added, and unreacted uracil was removed by extraction with H_2O. The solvent was dried and evaporated; trituration of the residue with EtOAc gave **12a** as a white powder, mp 212–214 °C (lit.[40] mp 216–217 °C). A small second crop of crystals was obtained from the EtOAc by preparative TLC (CH_2Cl_2–MeOH, 94:6). Total recovery was 0.905 g (84%). ^1H NMR ($CDCl_3$): δ 8.34 (br s, H3, 1), 7.92 (m, aromatic, 4), 7.53 (d, H6, 1), 7.27 (m, aromatic, 4), 5.61 (~dt, H3′, 1), 5.59 (dd, H5, 1), 4.74 (dd, H5′, 1), 4.68 (dd, H5″, 1), 4.54 (m, H4′, 1), 2.75 (dd, H2″, 1), 2.44 (s, CH_3, 3), 2.43 (s, CH_3, 3), 2.30 (dd, H2′, 1); $J_{2',2''}$ = 14.3, $J_{2',3'}$ = 6.6, $J_{2'',3'}$ = 2.0, $J_{3',4'}$ = 2.2, $J_{4',5'}$ = 3.2, $J_{4',5''}$ = 3.4, $J_{5',5''}$ = 12.3, $J_{5,6}$ = 8.2 Hz. ^{13}C NMR ($CDCl_3$): δ 166.08, 162.83, 150.14, 144.59, 138.84, 129.86, 129.47, 129.31, 126.53, 126.27, 102.92, 82.99, 74.62, 63.99, 38.18, 21.67. ^2H NMR (CH_2Cl_2): δ 6.32 (br s, D1′). MS (CI⁺):

(39) Andersen, W.; Dekker, C. A.; Todd, A. R. *J. Chem. Soc.* **1952**, 2721.

(40) Prystas, M.; Farkas, J.; Sorm, F. *Collect. Czech. Chem. Commun.* **1963**, *28*, 3140.

m/z 466 (M + H), 354, 137, 119, 113. UV (EtOH): λ_{max} (ϵ) 242 nm (33 600).

Compound **12b** was prepared in 92% yield as described for **12a**. The ^{1}H NMR spectrum was essentially identical with that of **12a** except that only residual signals were observed for H2′ and H2″. The ^{13}C spectrum showed weak multiplets at 38.0 (C2′) and 85.1 (C1′). ^{2}H NMR ($CHCl_3$): δ 6.38 (br s, D1′), 2.71 (br s, D2″), 2.28 (br s, D2′). MS (FAB$^+$): m/z 468 (M + H), 356, 119, 113.

Conversion of Deuteriated 2′-Deoxyuridines 12a–b to 2′-Deoxycytidines 14a–b. Method A. Compound **12a** (0.356 g, 0.767 mmol) in $CHCl_3$ (3.8 mL) was treated with purified $SOCl_2$ (0.91 mL, 11 mmol) and DMF (0.037 mL) at reflux for 1.25 h.[28] Volatile materials were removed by evaporation, and the residue was redissolved in CH_2Cl_2 and re-evaporated. The residue was purified by preparative TLC (CH_2Cl_2–MeOH, 97:3) to give 0.22 g of **13** and 0.098 g of recovered **12a**; the yield of 4-chloro-1-(1′-deuterio-3′,5′-di-*O*-*p*-toluoyl-2′-deoxyribosyl)-1*H*-pyrimidin-2-one (**13**) was 84% based on starting material consumed. Recrystallization from $CHCl_3/C_6H_6$ gave mp 175–175.5 °C. ^{1}H NMR ($CDCl_3$): δ 8.05 (d, H6, 1), 7.88 (m, aromatic, 4), 7.26 (m, aromatic, 4), 6.26 (d, H5, 1), 5.60 (m, H3′, 1), 4.72 (m, H4′,5′,5″, 3), 3.60 (dd, H2″, 1), 2.43 (s, CH_3, 3), 2.42 (s, CH_3, 3), 2.27 (dd, H2′, 1); $J_{2',2''}$ = 14.6, $J_{2',3'}$ = 6.5, $J_{2'',3'}$ = 1.7, $J_{3',4'}$, $J_{4',5'}$, $J_{4',5''}$ not determined, $J_{5,6}$ = 7.1 Hz. ^{13}C NMR ($CDCl_3$): δ 166.81, 166.00, 153.03, 144.64, 142,84, 129.79, 129.40, 129.27, 126.23, 105.16, 84.28, 74.86, 63.96, 39.32, 21.67. ^{2}H NMR ($CHCl_3$) δ 6.15 (br s, D1′). MS (FAB$^+$): m/z 484 (M + H), 354, 131, 119. HRMS (FAB$^+$): calcd for $C_{25}H_{23}D_1N_2ClO_6$ 484.1384, found 484.1389. UV (CH_3CN): λ_{max} (ϵ) 241 (39 900), 305 nm (7600).

Chloropyrimidine **13** (0.16 g, 0.33 mmol) was treated with methanolic ammonia (saturated at 0 °C) in a sealed heavy-wall vial at 50 °C for 10 h. The solvent was evaporated, and the residue was partitioned between H_2O and CH_2Cl_2. The aqueous layer was washed with CH_2Cl_2 and with Et_2O and evaporated. Preparative TLC (CH_2Cl_2–MeOH, 65:35) gave 1′-deuteriodeoxycytidine (**14a**) in 95% yield. Recrystallization from MeOH gave mp 195–196 °C (lit.[12a] mp 211–212 °C). ^{1}H NMR (D_2O): δ 7.83 (d, H6, 1), 6.05 (d, H5, 1), 4.45 (~dt, H3′, 1), 4.07 (m, H4′, 1), 3.86 (dd, H5′, 1), 3.77 (dd, H5″, 1), 2.44 (dd, H2″, 1), 2.28 (dd, H2′, 1); $J_{2',2''}$ = 14.1, $J_{2'',3'}$ = 4.0, $J_{2',3'}$ = 6.6, $J_{3',4'}$ = 3.8, $J_{4',5'}$ = 3.5, $J_{4',5''}$ = 5.3, $J_{5',5''}$ = 12.4, $J_{5,6}$ = 7.5 Hz. ^{13}C NMR ($CDCl_3$): δ 166.44, 157.74, 141.96, 96.56, 87.03, 86.22 (t, C1′, $^1J_{C,D}$ = 26 Hz), 70.99, 61.77, 39.55. ^{2}H NMR (H_2O): δ 6.06 (br s, D1′). MS (FAB$^+$): m/z 331 (M + H + glycerol), 229 (M + H), 112. UV (H_2O): λ_{max} (ϵ) 270 (9820), 230 nm (8720).

Method B. Compound **12a** (348 mg, 0.75 mmol) in CH_3CN (7.5 mL) was treated with triethylamine (2.4 mL), 1,2,4-triazole (1.16 gm, 16.9 mmol), and $POCl_3$ (280 mg, 1.83 mmol) for 3 h at ambient temperature. The mixture was poured into $CHCl_3$ (150 mL) containing 9 mL of triethylamine and washed with dilute $NaHCO_3$. The organic fraction was dried and evaporated; the residue was recrystallized from $CHCl_3$–EtOAc to give 115 mg of 4-(1,2,4-triazolyl)-1-(1′-deuterio-3′,5′-di-*O*-*p*-toluoyl-2′-deoxyribosyl)-1*H*-pyrimidin-2-one (**15**), mp 243–244 °C. An additional 132 mg was obtained by chromatography and recrystallization of the supernatant (combined yield 64%). ^{1}H NMR ($CDCl_3$): δ 9.20 (s, triazole H5, 1), 8.30 (d, H6, 1), 8.10 (s, triazole H3, 1), 7.95 (d, aromatic, 2), 7.85 (d, aromatic, 2), 7.30 (d, aromatic, 2), 7.20 (d, aromatic, 2), 6.92 (d, H5, 1), 5.65 (m, H3′, 1), 4.85 (m, H5′ and 5″, 2), 4.65 (m, H4′, 1), 3.20 (m, H2″, 1), 2.45 (s, CH_3, 3), 2.35 (s, CH_3, 3), 2.30 (m, H2′, 1). MS (CI$^+$): m/z 517 (M + H), 354, 164, 137, 119, 82. HRMS (FAB$^+$): calcd for $C_{27}H_{25}D_1N_5O_6$ 517.1944, found 517.1948.

A mixture of triazolide **15** (222 mg, 0.43 mmol), concentrated NH_4OH (1.6 mL), and dioxane (10 mL) was heated at 40 °C for 6 h and allowed to stand for 12 h at ambient temperature. The solvent was evaporated in vacuo, and the residue was partitioned between H_2O and CH_2Cl_2. The organic extracts were combined and dried to yield 168 mg (85%) of 1′-deuterio-2′-deoxy-3′,5′-di-*O*-*p*-toluoyl-2′-deoxycytidine (**16**) as an amorphous white solid. ^{1}H NMR ($CDCl_3$): δ 7.93 (d, aromatic, 2), 7.87 (d, aromatic, 2), 7.60 (d, H6, 1), 7.29 (d, aromatic, 2), 7.22 (d, aromatic, 2), 5.75 (d, H5, 1), 5.57 (m, H3′, 1), 4.64 (m, H5′, 5″, 2), 4.53 (m, H4′, 1), 2.83 (m, H2″, 1), 2.45 (s, CH_3, 3), 2.39 (s, CH_3, 3), 2.25 (m, H2′, 1). MS (CI$^+$): m/z 465 (M + H), 354, 329, 137, 119, 112, 82. HRMS (FAB$^+$): calcd for $C_{25}H_{25}D_1N_3O_6$ 465.1883, found 465.1881.

The toluoyl groups were removed with methanolic NH_3 as for compound **13** to give 59 mg (72%) of 1′-deuterio-2′-deoxycytidine (**14a**), mp 183–189 °C; the ^{1}H NMR was identical with material prepared by method A.

Method C. Compound **12b** (700 mg, 1.5 mmol) in dry pyridine (100 mL) under argon was mixed with diphenyl phosphorochloridate (1.3 mL, 4.2 equiv, 6.3 mmol) and 3-nitro-1,2,4-triazole (954 mg, 5 equiv, 7.5 mmol). The mixture was heated at 60 °C for 24 h. The solution was concentrated in vacuo, diluted with CH_2Cl_2, washed with $NaHCO_3$ and with H_2O, dried, and evaporated. The residue was chromatographed on TLC silica gel using a 0 → 2% MeOH/CH_2Cl_2 gradient followed by preparative TLC on silica gel eluted successively with 2%, 4%, 6%, and 8% MeOH/CH_2Cl_2. Elution of the product band with 20% MeOH/CH_2Cl_2 and trituration of the material with cold EtOH gave 566 mg (67%) of 4-(3-nitro-1,2,4-triazolyl)-1-(1′,2′,2″-trideuterio-3′,5′-di-*O*-*p*-toluoyl-2′-deoxyribosyl)-1*H*-pyrimidin-2-one (**17**), mp 223–224 °C. In addition, 91 mg of unreacted **12b** (13%) was removed from the reaction. Compound **17**. ^{1}H NMR ($CDCl_3$): δ 9.29 (s, triazole H5, 1), 8.47 (d, H6, 1), 7.97 (~d, aromatic, 2), 7.83 (~d, aromatic, 2), 7.29 (~d, aromatic, 2), 7.21 (~d, aromatic), 6.95 (d, H5, 1), 5.64 (d, H3′, 1), 4.88 (dd, H5′, 1), 4.7 (m, H4′, 1), 4.68 (dd, H5″, 1), 2.45 (s, CH_3, 3), 2.38 (s, CH_3, 3); $J_{3',4'}$ = 2.1, $J_{4',5'}$ = 2.7, $J_{4',5''}$ = 3.6, $J_{5',5''}$ = 12.3, $J_{5,6}$ = 7.2 Hz. ^{13}C NMR ($CDCl_3$): δ 166.0, 165.9, 158.4, 153.4, 147.2, 144.7, 144.5, 129.8, 129.4, 129.3, 126.3, 126.1, 94.0, 88.8 (weak multiplet), 84.6, 74.6, 63.8, 39.0 (weak multiplet), 21.7, 21.6. ^{2}H NMR ($CHCl_3$): δ 6.32 (br s, D1′), 3.18 (br s, D2″), 2.32 (br s, D2′). MS (FAB$^+$): m/z 564 (M + H), 356, 119. UV (EtOH): λ_{max} (ϵ) 242 (16 700), 323 nm (2200). Anal. Calcd for $C_{27}H_{21}D_3N_6O_8$: C, 57.57; H/D, 4.29; N, 14.92. Found: C, 57.99; H/D, 4.02; N, 14.49.

Compound **17** was converted to 1′,2′,2″-trideuterio-2′-deoxycytidine (**14b**) by ammonolysis at 40 °C for 24 h. Purification by C_{18} reverse-phase column chromatography (30% acetone/H_2O) followed by C_2 preparative TLC (1% MeOH/CH_2Cl_2) gave 135 mg (67%) of **14b**, mp 210–212 °C. ^{1}H NMR (D_2O): δ 7.83 (d, H6, 1), 6.03 (d, H5, 1), 4.43 (d, H3′, 1), 4.07 (m, H4′, 1), 3.85 (dd, H5′, 1), 3.77 (dd, H5″, 1), 2.42 (m, residual H2′, 0.14), 2.28 (m, residual H2″, 0.05); $J_{3',4'}$ = 3.8, $J_{4',5'}$ = 3.5, $J_{4',5''}$ = 5.2, $J_{5',5''}$ = 12.5, $J_{5,6}$ = 7.6 Hz. ^{13}C NMR (D_2O): δ 166.4, 157.5, 143.4, 97.0, 87.4, 86.9 (weak multiplet), 71.6, 62.5, 40.1 (weak multiplet). ^{2}H NMR (H_2O): δ 6.00 (br s, D1′), 2.19 (br s, D2″), 2.04 (br s, D2′). MS (FAB$^+$): m/z 231 (M + H), 112. UV (H_2O): λ_{max} (ϵ) 269 nm (7400).

6-Chloro-9-(1′-deuterio- and 1′,2′,2″-trideuterio-3′,5′-di-*O*-*p*-toluoyl-2′-deoxyribosyl)purine (18a–b).[30b] 6-Chloropurine (0.316 g, 2.06 mmol) was added to a suspension of 49 mg (2.05 mmol) of NaH in 50 mL of dry CH_3CN. The mixture was stirred until most of the chloropurine had dissolved (ca. 20 min) and then treated with chloro sugar **9a** (0.4 g, 1.03 mmol), which was added all at once. After 4 h the reaction was quenched with MeOH (0.2 mL). The mixture was filtered; the filtrate was evaporated, taken up in CH_2Cl_2 and filtered again. The filtrate was chromatographed on silica gel (MeOH/CH_2Cl_2 mixtures up to 1.5% MeOH); impure fractions were further purified by preparative TLC (CH_2Cl_2–MeOH, 94:6) to give 0.278 g (53%) of **18a**, which was crystallized from EtOH, mp 108–109 °C (lit.[30b] mp 107–109 °C). ^{1}H NMR ($CDCl_3$) δ 8.68 (s, H8, 1), 8.30 (s, H2, 1), 7.94 (m, aromatic, 4), 7.25 (m, aromatic, 4), 5.85 (dt, H3′, 1), 4.80 (m, H4′, 1), 4.67 (m, H5′,5″, 2), 3.18 (dd, H2′,1), 2.89 (dd, H2″, 1), 2.45 (s, CH_3, 3), 2.41 (s, CH_3, 3); $J_{2',2''}$ = 14.2, $J_{2',3'}$ = 6.3, $J_{2'',3'}$ = 2.2, $J_{3',4'}$, $J_{4',5'}$, $J_{4',5''}$, $J_{5',5''}$ not determined. ^{13}C NMR ($CDCl_3$): δ 166.1, 165.94, 152.04, 151.36, 151.14, 144.63, 144.29, 143.46, 132.30, 129.81, 129.57, 129.30, 126.53, 126.34, 85.1 (C1′), 83.41, 74.97, 63.73, 37.78, 21.63. ^{2}H NMR (H_2O): δ 6.55 (br s, D1′). MS (CI$^+$): m/z 508 (M + H), 354, 155, 137, 119. UV (EtOH): λ_{max} 246, 264 nm (sh).

6-Chloropurine (632 mg, 4.12 mmol) was treated with 800 mg of **9b** (2.06 mmol, mp, 120–121 °C) as described above to give 636 mg of **18b** (60%), mp 112–113 °C. The ^{1}H NMR spectrum was the same as that of **18a** except that the signals for H2′ and H2″ were missing and H3′ was a broad singlet. The ^{13}C spectrum was missing the signal at 37.78 (C2′). ^{2}H NMR ($CHCl_3$): δ 6.57 (br s, D1′), 3.17 (br s, D2′), 2.87 (br s, D2″). MS (FAB$^+$): m/z 510 (M + H), 356, 119.

1′-Deuterio- (19a) and 1′,2′,2″-Trideuterio-2′-deoxyadenosine (19b). Compound **18a** (0.214 g, 0.42 mmol) was

converted to **19a** by ammonolysis (100 °C for 18 h). The solvent was evaporated; the residue was dissolved in water (10 mL) and washed with CH_2Cl_2 and Et_2O. The water was evaporated, and the residue recrystallized from hot H_2O to yield 0.079 g (70%) of 1'-deuterio-2'-deoxyadenosine (**19a**) as the hydrate, mp 185–188 °C (lit.[39] mp 189–190 °C). ^1H NMR (D_2O): δ 8.22 (s, H8, 1), 8.06 (s, H2, 1), 4.65 (m, H3', 1), 4.19 (m, H4', 1), 3.87 (dd, H5', 1), 3.80 (dd, H5'', 1), 2.77 (dd, H2', 1,), 2.56 (dd, H2'', 1); $J_{2',2''}$ = 14.0, $J_{2',3'}$ = 6.1, $J_{2'',3'}$ = 3.4, $J_{3',4'}$ not determined, $J_{4',5'}$ = 3.2, $J_{4',5''}$ = 4.3, $J_{5',5''}$ = 12.7 Hz. ^{13}C NMR (D_2O): δ 155.72, 152.70, 148.53, 140.56, 119.16, 87.89, 71.72, 62.15, 40.54. ^2H NMR (H_2O): δ 6.16 (br s, D1'). MS (CI^+): *m/z* 253 (M + H), 164, 136, 135, 118. MS (FAB^+): *m/z* 253 (M + H). UV (H_2O): λ_{max} (ε) 258 nm (15500).

Ammonolysis of compound **18b** (550 mg, 1.08 mmol) gave 207 mg (75%) of **19b**; mp 184–187 °C; 55 mg (19%) of the 6-methoxy purine analogue was also isolated. **19b**: The ^1H NMR differed from that of **18a** in that the signals for H2' and H2'' were missing; H3' appeared as a doublet ($J_{3',4'}$ = 2.7 Hz). In the ^{13}C spectrum the signal at 40.53 was replaced by a weak multiplet at 39.0. ^2H NMR (H_2O): δ 6.09 (br s, D1'), 2.48 (br s, D2'), 2.29 (br s, D2''). MS (FAB^+): *m/z* 255 (M + H), 136, 120.

1'-Deuterio-2'-deoxyguanosine (22a). 2-Amino-6-chloropurine (1.16 g, 6.84 mmol) in DMF (20 mL) was treated with di-*n*-butylformamide diethyl acetal[32b] (2.36 g, 9.33 mmol) for 6 h at ambient temperature. The solvent was evaporated; the residue was extracted with hot EtOAc. Crystallization followed by preparative TLC (10% MeOH/CH_2Cl_2) gave 1.75 g (58%) of 6-chloro-2-(*N*-((di-*n*-butylamino)methylene)amino)purine (**20**), mp 153–155 °C after recrystallization from CH_3CN. ^1H NMR ($CDCl_3$): δ 14.39 (v br s, H9, 1), 8.79 (s, CH=N, 1), 8.08 (s, H8, 1), 3.58 (t, NCH_2, 2), 3.38 (t, NCH_2, 2), 1.65 (m, CH_2, 4), 1.36 (m, CH_2, 4), 0.97 (t, CH_3, 3), 0.92 (t, CH_3, 3). ^{13}C NMR ($CDCl_3$): δ 162.06, 158.49, 154.34, 150.46, 126.95, 52.11, 45.58, 31.08, 29.05, 20.24, 19.72, 13.77, 13.61. MS (CI^+): *m/z* 309 (M + H), 273. UV (EtOH): λ_{max} (ε) 310 nm (20500), 282 (23500), 238 (14700). Anal. Calcd for $C_{14}H_{21}N_6Cl$: C, 27.21; H, 54.45; N, 6.85. Found: C, 27.36; H, 54.17; N, 6.83.

A warm solution of **20** (0.523 g, 1.7 mmol) in CH_3CN (25 mL) was treated with 40 mg (1.7 mmol) of NaH. The resulting anion was treated with 0.600 g (1.54 mmol) of solid **9a** at room temperature, followed after 10 min by 4.62 mmol of sodium pyridine-2-carbaldoximate (prepared from equal molar quantities of the oxime and NaH in CH_3CN (15 mL). After 2 h the reaction was quenched with MeOH (1 mL) followed by 0.137 mL of acetic acid. The solvent was evaporated, and the residue was partitioned between H_2O and CH_2Cl_2. The organic layer was washed with H_2O, dried, evaporated, and chromatographed on silica gel (CH_2Cl_2–MeOH, up to 3% MeOH) to give the following components, which are described in their order of elution. A substance tentatively assigned as 3,7-[bis(1'-deuterio-3',5'-di-*O*-*p*-toluoyl-2'-deoxyribosyl)]-2-[*N*-[(di-*n*-butylamino)methylene]amino]-3*H*,7*H*-purin-6-one was further purified by reverse-phase TLC (85% acetone/H_2O). ^1H NMR ($CDCl_3$): δ 8.85 (s, CH=N, 1), 7.92 (m, aromatic and H8, 9), 7.22 (m, aromatic, 8), 5.96 (m, H3'a, 1), 5.64 (m, H3'b, 1), 4.71 (m, H4'b, 5'a, 5''a, 5'b, 5''b, 5), 4.51 (m, H4'a, 1), 3.64 (m, H2a, NC*H'*H'', 2), 3.46 (m, NCH'*H''*, 1), 3.36 (t, NCH_2, 2), 3.02 (dd, H2''b, 1), 2.59 (dd, H2'b, 1), 2.42 (s + m, CH_3 and H2''a, 7), 2.37 (s, CH_3, 3), 2.34 (s, tolyl CH_3, 3), 1.66 (m, CH_2 and H_2O, 4), 1.33 (m, CH_2, 4), 0.93 (m, CH_3, 6); $J_{2b,2''b}$ = 14.4, $J_{2'b,3'b}$ = 6.9, $J_{2''b,3'b}$ = 2.5 Hz, other *J* values not determined. MS (FAB^+): *m/z* 996 (M + H), 291, 119.

Another substance was tentatively identified as 7-(1'-deuterio-3',5'-di-*O*-*p*-toluoyl-2'-deoxyribosyl)-2-(*N*-((di-*n*-butylamino)methylene)amino)-7*H*-purin-6(1*H*)-one. ^1H NMR ($CDCl_3$): δ 8.80 (s, CH=N, 1), 8.51 (br s, H3, 1), 8.07 (s, H8, 1), 7.94 (m, aromatic, 4), 7.24 (m, aromatic, 4), 5.68 (m, H3', 1), 4.70 (m, H5',5'', 2), 4.63 (m, H4', 1), 3.47 (t, NCH_2, 2), 3.33 (t, NCH_2, 2), 2.93 (dd, H2'', 1), 2.78 (dd, H2', 1), 2.43 (s, CH_3, 3), 2.39 (s, CH_3, 3), 1.61 (m, CH_2, 4), 1.35 (m, CH_2, 4), 0.96 (m, CH_3, 6); $J_{2,2''}$ = 14.3, $J_{2',3'}$ = 6.6, $J_{2'',3'}$ = 2.4, other *J* values not determined. ^{13}C NMR ($CDCl_3$): δ 166.24, 165.98, 158.02, 156.57, 154.84, 144.36, 144.05, 140.52, 129.88, 129.69, 129.29, 83.21, 74.89, 64.15, 52.21, 45.62, 40.03, 31.02, 29.00, 21.65, 20.15, 19.76, 13.78, 13.65. MS (FAB^+): *m/z* 644 (M + H), 409, 291, 119. UV (EtOH): λ_{max} 238, 294 nm. A_{238}/A_{294} = 1.151/0.652.

The next major component eluted was the desired 9β-(1'-deuterio-3',5'-di-*O*-*p*-toluoyl-2'-deoxyribosyl)-2-(*N*-((di-*n*-butylamino)methylene)amino)-9*H*-purin-6(1*H*)-one (**21**), which was further purified by reverse-phase chromatography (66% acetone/water) to give 0.256 g (26%). ^1H NMR ($CDCl_3$): δ 8.92 (s, CH=N, 1), 8.70 (br s, H1, 1), 7.88 (m, aromatic, 4), 7.69 (s, H8, 1), 7.23 (m, aromatic, 4), 6.00 (m, H3', 1), 4.63 (m, H5',5'',4', 3), 3.46 (t, NCH_2, 2), 3.40 (t, NCH_2, 2), 3.27 (dd, H2', 1), 2.69 (dd, H2'', 1), 2.44 (s, CH_3, 3), 2.39 (s, CH_3, 3), 1.58 (m, CH_2, 4), 1.32 (m, CH_2, 4), 0.95 (t, CH_3, 3), 0.89 (t, CH_3, 3); $J_{2',2''}$ = 14.0 Hz, $J_{2',3'}$ = 7.0 Hz, $J_{2'',3'}$ = 4.7 Hz. ^{13}C NMR ($CDCl_3$): δ 166.91, 165.93, 157.63, 156.83, 149.76, 144.37, 143.94, 137.07, 129.65, 129.51, 129.16, 129.11, 126.60, 126.48, 121.23, 81.90, 74.29, 63.47, 51.97, 45.59, 36.74, 30.91, 28.88, 21.60, 20.06, 19.59, 13.68, 13.55. ^2H NMR (CH_2Cl_2): δ 6.28 (br s, D1'). MS (FAB^+): *m/z* 644 (M + H), 291, 119. HRMS (FAB^+): calcd for $C_{35}H_{42}D_1N_6O_6$ 644.3305, found 644.3307. UV (EtOH): λ_{max} (ε) 241 (49900), 265 (sh), 272 (sh), 297 (25300).

The final major component eluted was tentatively assigned as the 3-isomer, 3-(2'-deoxy-1'-deuterio-3',5'-di-*O*-*p*-toluoyl-β-D-*erythro*-pentofuranosyl)-2-*N*-(((di-*n*-butylamino)methylene)amino)-3*H*-purin-6(7*H*)-one. ^1H NMR ($CDCl_3$): δ 8.92 (s, CH=N, 1), 7.96 (m, aromatic, 4), 7.85 (s, H8, 1), 7.21 (m, aromatic, 4), 6.05 (m, H3', 1), 4.89 (dd, H5', 1), 4.75 (dd, H5'', 1), 4.54 (m, H4', 1), 3.82 (dd, H2', 1), 3.64 (m, NC*H'*H'', 1), 3.48 (m, NCH'*H''*, 1), 3.39 (t, NCH_2, 2), 2.44 (dd, H2'', 1), 2.43 (s, tolyl CH_3, 3), 2.38 (s, tolyl CH_3, 3), 1.64 (m, CH_2, 4), 1.35 (m, CH_2, 4), 0.94 (m, CH_3, 6); $J_{2',2''}$ = 13.9 Hz, $J_{2',3'}$ = 7.5 Hz, $J_{2'',3'}$ = 3.7 Hz, $J_{5',4'}$ = 5.2 Hz, $J_{5',5''}$ = 11.6 Hz, $J_{5'',4'}$ = 6.0 Hz. The NMR spectrum was also obtained in DMSO-d_6. The resonances were generally the same in chemical shift and coupling, although resolution was not as good. The $H_{7/9}$ resonance was found at 13.22 ppm. ^{13}C NMR ($CDCl_3$): δ 166.44, 165.92, 163.56, 159.16, 156.79, 147.67, 143.93, 143.49, 139.65, 129.87, 129.71, 129.07, 128.92, 127.32, 127.10, 114.31, 85.17, 81.82, 75.82, 64.37, 52.42, 46.05, 35.02, 31.15, 29.06, 21.62, 20.20, 19.83, 13.73, 13.68. ^2H NMR (CH_2Cl_2): δ 7.49 (br s, D1'). MS (positive ion FAB): *m/z* 644 (M + H), 291, 119. Several other components were present which were not examined in detail.

Compound **21** (0.200 g, 0.311 mmol) was heated at 60 °C in a sealed vial with methanolic NH_3 (8 mL), prepared by NH_3 saturation of MeOH at 0 °C. The solvent was evaporated and the residue partitioned between CH_2Cl_2 and H_2O. The aqueous fraction was washed with CH_2Cl_2 and Et_2O to give after concentration and crystallization 0.076 g (91%) of 1'-deuterio-2'-deoxyguanosine (**22a**), mp >300 °C (lit.[31f] mp >300 °C). ^1H NMR (D_2O): δ 7.97 (s, H8, 1), 4.62 (m, H3', 1), 4.12 (m, H4', 1), 3.82 (dd, H5', 1), 3.75 (dd, H5'', 1), 2.77 (dd, H2', 1), 2.50 (dd, H2'', 1); $J_{2',2''}$ = 14.1, $J_{2',3'}$ = 6.2, $J_{2'',3'}$ = 3.5, $J_{3',4'}$ not determined, $J_{4',5'}$ = 3.6, $J_{4',5''}$ = 4.7, $J_{5',5''}$ = 12.5 Hz. ^{13}C NMR (DMSO-d_6, DMSO standard 39.5 ppm): δ 156.79, 153.66, 150.82, 135.20, 116.65, 87.54, 82.28 (1'-CD), 70.72, 61.70, (the signal for C-2' is presumably hidden by the DMSO heptet at 40.12 to 38.87 ppm). ^2H NMR (H_2O): δ 6.05 (br s, D1'). MS (FAB^+): *m/z* 269 (M + H). UV (H_2O): λ_{max} (ε) 252 (13600).

1',2',2''-Trideuterio-2'-deoxyguanosine. 2-Amino-6-chloropurine (0.52 g, 3.1 mmol) was treated with NaH (74 mg, 3.1 mmol) in warm CH_3CN (25 mL) under nitrogen; the mixture was cooled, and 0.80 g of **9b** (2.04 mmol) was added as a solid. After 30 min CH_2Cl_2 (100 mL) was added, and the salts were filtered off. The organic solution was washed with H_2O, dried, and evaporated. Column chromatography on TLC grade silica gel (CH_2Cl_2, 0 → 1% MeOH/CH_2Cl_2) followed by preparative TLC (5% MeOH/CH_2Cl_2) gave 649 mg (61%) of 2-amino-6-chloro-9-(1',2',2''-trideuterio-3',5'-di-*O*-*p*-toluoyl-2'-deoxyribosyl)purine (**23**), mp 184–186 °C (lit.[12b] mp 176–178 °C). ^1H NMR ($CDCl_3$): δ 7.98 (d, aromatic, 2), 7.90 (s, H8, 1), 7.88 (d, aromatic, 2), 7.54 (d, aromatic, 2), 7.43 (d, aromatic, 2), 5.82 (d, H3', 1), 5.16 (br s, NH_2, 2), 4.85 (dd, H5', 1), 4.66 (m, H5'' and H4', 2), 2.45 (s, CH_3, 3), 2.42 (s, CH_3, 3); $J_{3',4'}$ = 1.9, $J_{4',5'}$ = 5.8, $J_{4',5''}$ not determined, $J_{5',5''}$ = 13.1 Hz. ^{13}C NMR ($CDCl_3$): δ 166.3, 165.9, 159.0, 151.8, 144.5, 144.2, 140.5, 129.8, 129.6, 129.3, 126.6, 126.5, 82.9, 75.0, 63.8, 21.7. ^2H NMR ($CHCl_3$): δ 6.37 (br s, D1'), 3.17 (br s, D2'), 2.74 (br s, D2''). MS (FAB^+): *m/z* 525 (M + H), 356, 119.

Compound **23** (211 mg, 0.40 mmol) was treated for 1 h at 40 °C with sodium β-cyanoethoxide [prepared from 426 mg (6.0 mmol) of β-cyanoethanol and 156 mg (6.5 mmol) of NaH in THF (60 mL)]. The reaction was quenched with 0.4 mL (6.99 mmol)

of HOAc. The solvents were removed in vacuo. The residue was taken up in H_2O (40 mL), washed with Et_2O and CH_2Cl_2, concentrated, and chromatographed on Dowex 1-X8 (100-200 mesh, formate form) with elution by H_2O to yield 64 mg (56%) of 1′,2′,2″-d_3-2′-deoxyguanosine (**22b**) as a white powder, mp >300 °C. ^1H NMR (d_6-DMSO): δ 7.89 (s, H8, 1H), 6.65 (br s, NH_2, 2), 4.33 (d, H3′, 1), 3.80 (m, H4′, 1), 3.55 (dd, H5″, 1), 3.47 (dd, H5′, 1); $J_{3',4'}$ = 2.4, $J_{4',5'}$ = 4.3, $J_{4',5''}$ = 4.4, $J_{5',5''}$ = 11.5 Hz. ^{13}C NMR (d_6-DMSO) δ 157.9, 154.5, 150.9, 135.1, 116.7, 87.6, 82.1 (weak multiplet), 70.7, 61.8. ^2H NMR (H_2O): δ 6.06 (br s, D1′), 2.57 (br s, D2′), 2.30 (br s, D2″). MS (FAB$^+$): m/z 271 (M + H), 152, 120. UV (H_2O): λ_{max} (ϵ) 252 (13000), 272 nm (sh).

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Registry No. 1, 5336-08-3; 2, 78138-89-3; **3a**, 34371-14-7; **3b**, 131132-17-7; **4a**, 83159-91-5; **4b**, 131132-08-6; α-**5a**, 131132-08-6; β-**5a**, 131132-25-7; α-**5b**, 131132-09-7; β-**5b**, 131232-93-4; **6a**, 131131-90-3; **6b**, 131132-10-0; α-**7a**, 131131-91-4; β-**7a**, 131132-26-8; α-**7b**, 131132-11-1; β-**7b**, 131132-28-0; α-**8a**, 131131-92-5; β-**8a**, 131132-27-9; α-**8b**, 131132-12-2; β-**8b**, 131132-29-1; **9a**, 131131-93-6; **9b**, 131132-13-3; **10a**, 131131-94-7; **10b**, 131132-14-4; **11a**, 131131-95-8; **11b**, 131132-15-5; **12a**, 131131-96-9; **12b**, 131132-16-6; **13**, 131131-97-0; **14a**, 131131-98-1; **14b**, 131132-18-8; **15**, 131131-99-2; **16**, 131132-00-8; **17**, 131132-01-9; **18a**, 131132-02-0; **18b**, 131132-19-9; **19a**, 131132-03-1; **19b**, 131132-20-2; **20**, 131132-04-2; **21**, 131132-05-3; **21** N^3 isomer, 131132-24-6; **21** N^7 isomer, 131132-23-5; **22a**, 131132-06-4; **22b**, 131132-22-4; **23**, 131132-07-5; thymine, 65-71-4; uracil, 66-22-8; 6-chloropurine, 87-42-3; 2-amino-6-chloropurine, 10310-21-1; 3,7-[bis(1′-deuterio-3′,5′-di-*O*-*p*-toluoyl-2′-deoxyribosyl)]-2-[*N*-[(di-*n*-butylamino)methylene]-amino]-3*H*,7*H*-purin-6-one, 131132-21-3; sodium 2-cyanoethoxide, 131513-78-5.

Supplementary Material Available: ^1H NMR spectra of compounds **5a**, **5b**, **13**, **15**, **16**, and **21** (6 pages). Ordering information is given on any current masthead page.

Synthesis of the Phthalide Isoquinoline Alkaloids (-)-Egenine, (-)-Corytensine, and (-)-Bicuculline by Asymmetric Carbonyl Addition of Chiral Dipole-Stabilized Organometallics

Kathleen S. Rein and Robert E. Gawley*

Department of Chemistry, University of Miami, Coral Gables, Florida 33124

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The asymmetric addition of metalated [(methylenedioxy)isoquinolyl]oxazolines is 100% selective for the erythro (α-hydroxybenzyl)isoquinoline diastereomers, with 2:1 selectivity of the two possible erythro stereoisomers. Enrichment to 100% ee after removal of the auxiliary and conversion to (-)-bicuculline and (+)-bicuculline diol establish the absolute configuration of the major addition product. Inversion of the C-9 hydroxyl affords entry into the threo series as well. The asymmetric carbonyl addition was used to synthesize, for the first time, the phthalide-isoquinoline hemiacetals corytensine and egenine, confirming the previously assigned structures and absolute configurations, and establishing the identity of egenine with decumbensine and of corytensine with epi-α-decumbensine.

The phthalide-isoquinoline alkaloids are a class of tetracyclic (α-hydroxybenzyl)isoquinoline lactones that are usually oxygenated on carbons 6, 7, 4′, and 5′.[1] One of the more important members of this class is bicuculline, **1**, which is of interest as a $GABA_A$ antagonist.[2] Its presumed biosynthetic precursor is the hemiacetal egenine, **2**.[3] Bicuculline and egenine both possess the erythro relative configuration at positions 1 and 9; bicuculline has been isolated as either enantiomer and as the racemate[1] (although only the (+) enantiomer is active as a $GABA_A$ antagonist[2b]), while egenine was isolated as the (+) enantiomer.[3] The threo diastereomers of bicuculline and egenine are the lactone (-)-capnoidine, **3** (and its enantiomer (+)-adlumidine),[1] and the hemiacetal corytensine, **4**, isolated as the (+) enantiomer.[4] In 1988, two (α-hydroxybenzyl)isoquinoline alkaloids were isolated from *Corydalis decumbens* (Thunb.) Pers. (Papaveraceae), named decumbensine and epi-α-decumbensine, and assigned structures **5** and **6**, respectively.[5] In 1989, Rozwadowska synthesized compounds **5** and **6** and found that spectra of the synthetic material did not match the published spectra of decumbensine and epi-α-decumbensine.[6] In this paper, we present evidence that decumbensine and egenine are identical and that epi-α-decumbensine and corytensine are identical.[7]

For the past several years, the asymmetric alkylation of isoquinolines, α to nitrogen, has been a topic of current interest.[8] The seminal contribution to this field was Meyers' asymmetric alkylation of chiral formamidines.[8a]

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