Table V

	Syn/anti ratio	Yield, g
Not degassed	2.65	0.513
	2.62	0.481
Degassed (2 cycles)	2.43	0.533
	2.46	0.524
Degassed (5 cycles)	2.44	0.534
	2.38	0.535

centrated to the residual solid which had dissolved. The *residue* was then weighed.

Isolation of the Anti Dimer. Acenaphthylene (10 g) was dissolved in 50 mL of *p*-OHC-C_6H_4-Br (20 mol %)/benzene and irradiated (*without* prior degassing) for 25 h. The crude product which had precipitated was isolated, washed with three portions (500 mL total) of hot cyclohexane, and recrystallized from benzene as white needles: mp 301–302 °C (lit. mp 306–307 °C);[9] UV (cyclohexane) 219 (ϵ 6.56 × 10^4), 225 nm (ϵ 1.11 × 10^5).

Isolation of the Syn Dimer. A solution of acenaphthylene (10 g) in methanol (50 mL) was degassed and irradiated for 25 h. The crude product which precipitated was isolated, and a portion of it was recrystallized from cyclohexane as white prisms: mp 232–234 °C (lit. mp 232–234 °C);[9] UV (cyclohexane) 219 (ϵ 1.10 × 10^5), 225 nm (ϵ 4.99 × 10^4).

Control. Photostability of Acenaphthylene Photodimers. The pure anti dimer (1.0 g, see above) was added to a sufficient quantity of methanol so that the final volume was 10 mL, and the resulting solid/liquid mixture was degassed and irradiated for 15 h in the usual fashion. Ultraviolet analysis of the insoluble "product" gave a syn/anti dimer ratio of 0.18.

The pure syn dimer was treated analogously and gave a syn/anti ratio of 4.37.

Control. Syn/Anti Ratios as a Function of Sample Degassing. In all of the reactions previously described, the reaction mixtures were degassed by *two* freeze–pump cycles. That two cycles are sufficient is indicated by the following study. Six reaction mixtures were prepared, each containing 1.0 g of acenaphthylene in 1.0 mol % of bromobenzene in methanol (total volume 10 mL). Two reaction mixtures were *not* degassed, two were degassed with *two* freeze (liquid N_2)–pump–thaw cycles, and two were degassed with *five* freeze–pump–thaw cycles. After irradiation and product analysis in standard fashion, the syn/anti ratios and dimer yields were obtained (Table V).

Registry No.—**1**, 208-96-8; *syn*-**1** photodimer, 15065-28-8; *anti*-**1** photodimer, 14620-98-5.

References and Notes

(1) D. O. Cowan and R. L. Drisko, *Tetrahedron Lett.*, 1255 (1967).
(2) D. O. Cowan and R. L. Drisko, *J. Am. Chem. Soc.*, **89**, 3068 (1967).
(3) D. O. Cowan and R. L. E. Drisko, *J. Am. Chem. Soc.*, **92**, 6281 (1970).
(4) D. O. Cowan and R. L. E. Drisko, *J. Am. Chem. Soc.*, **92**, 6286 (1970).
(5) D. O. Cowan and J. C. Koziar, *J. Am. Chem. Soc.*, **96**, 1229 (1974).
(6) D. O. Cowan and J. C. Koziar, *J. Am. Chem. Soc.*, **97**, 249 (1975).
(7) J. C. Koziar and D. O. Cowan, *J. Am. Chem. Soc.*, **98**, 1001 (1976).
(8) S. P. McGlynn, R. Sunseri, and N. Christodouleas, *J. Chem. Phys.*, **37**, 1818 (1962).
(9) K. Dziewonski and C. Pasehalski, *Ber., Dtsch. Chem. Ges.*, **46**, 86 (1913).
(10) I. M. Hartmann, W. Hartmann, and G. O. Schenck, *Chem. Ber.*, **100**, 3146 (1967).
(11) "CRC Handbook of Chemistry and Physics", 50th ed, 1969–1970, p E-63.
(12) *Natl. Bur. Stand. (U.S.) Circ.*, **No. 514** (1951).

Nucleosides. 108. Ribo–Xylo Interconversions of 6,5′-Cyclopyrimidine Nucleosides via Autoxidation and Retro-Aldol Reactions[1,2]

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The 5′*S* and 5′*R* epimers of 6,5′-cyclouridine undergo autoxidation to 5′-oxo-6,5′-cyclouridine when treated with oxygen and 1 N NaOH. 5′-Oxo-6,5′-cyclouridine is stable in 1 N NaOH, but under less strongly alkaline conditions, e.g., ethanolic ammonia, it undergoes 3′ epimerization to give 6,5′-cyclo-5′-oxo-1-(β-D-xylofuranosyl)uracil, probably via formation and recyclization of a pyrimido[1,6-*c*][1,3]oxazine intermediate generated by retro-aldol cleavage. The 5′-carbonyl group of 5′-oxo-6,5′-cyclouridine is predominately hydrated in aqueous systems, whereas the 5′-oxo-xylo isomer exists as the keto form under the same conditions. These ribo–xylo epimers consequently show large differences in ultraviolet spectral properties in water that are useful in monitoring the retro-aldol equilibrium reaction. Similar differences in the UV spectra of hydrated orotaldehyde (261 nm) and anhydrous orotaldehyde (300 nm) were noted. Reduction of 5′-oxo-6,5′-cyclouridine with sodium cyanoborohydride in acetic acid affords only 6,5′(*S*)-cyclouridine. Similar reduction of the 5′-oxo-xylo nucleoside affords both 5′*S* and 5′*R* epimers of 6,5′-cyclo-1-(β-D-xylofuranosyl)uracil in a ratio of 5:1, possibly indicating that the 5′*R*-xylo isomer is formed via participation of the 3′-hydroxyl group. The identity of each xylo 5′ epimer was established from NMR spectra and by the ready formation of a 3′,5′-*O*-isopropylidene derivative of the 5′*S* epimer.

Nucleosides and nucleotides restricted to one type of conformation, but retaining a full complement of hydrogen-bonding sites, are useful for probing the conformational factors that affect the specificities of the enzymes of nucleic acid metabolism.[3] In this regard, we have previously reported[4] the synthesis of the 5′*R* and 5′*S* epimers of 6,5′-cyclouridine (**1** and **2**, Scheme I). These nucleosides are fixed in the anti conformational range, and the orientations of the 5′-hydroxyl groups correspond approximately to the gauche–trans and trans–gauche $C_{4',5'}$ rotamers, respectively, of unrestricted nucleosides.

In addition to their potential as biochemical tools, 6,5′-cyclonucleosides are interesting from a chemical viewpoint because the allylic character of C-5′ enhances the reactivity of that position relative to ordinary nucleosides. For example, derivatives of **1** and **2** in which the 5′-hydroxyl groups are protected undergo base-catalyzed epimerization at C-5′ via a mechanism involving 5′-carbanion intermediates.[4] We now wish to report that 6,5′-cyclopyrimidine nucleosides with unsubstituted 5′-hydroxyl groups readily undergo base-catalyzed autoxidation and that the resulting 5′-oxo nucleosides can rearrange to give their D-xylo epimers.

The first example of autoxidation of a 6,5′-cyclopyrimidine nucleoside was encountered during the synthesis of inter-

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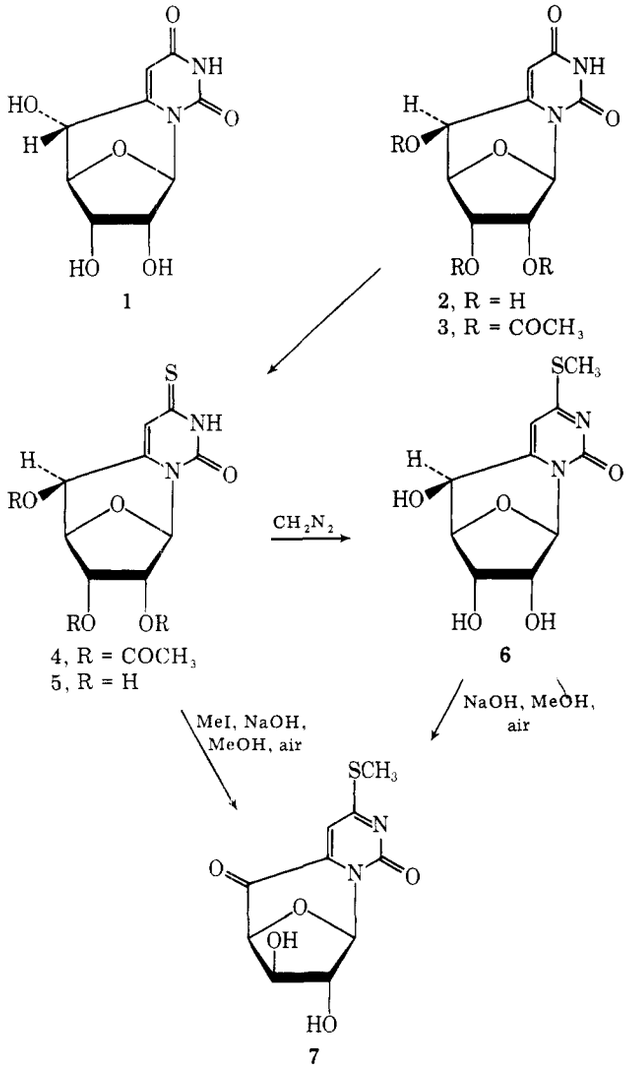
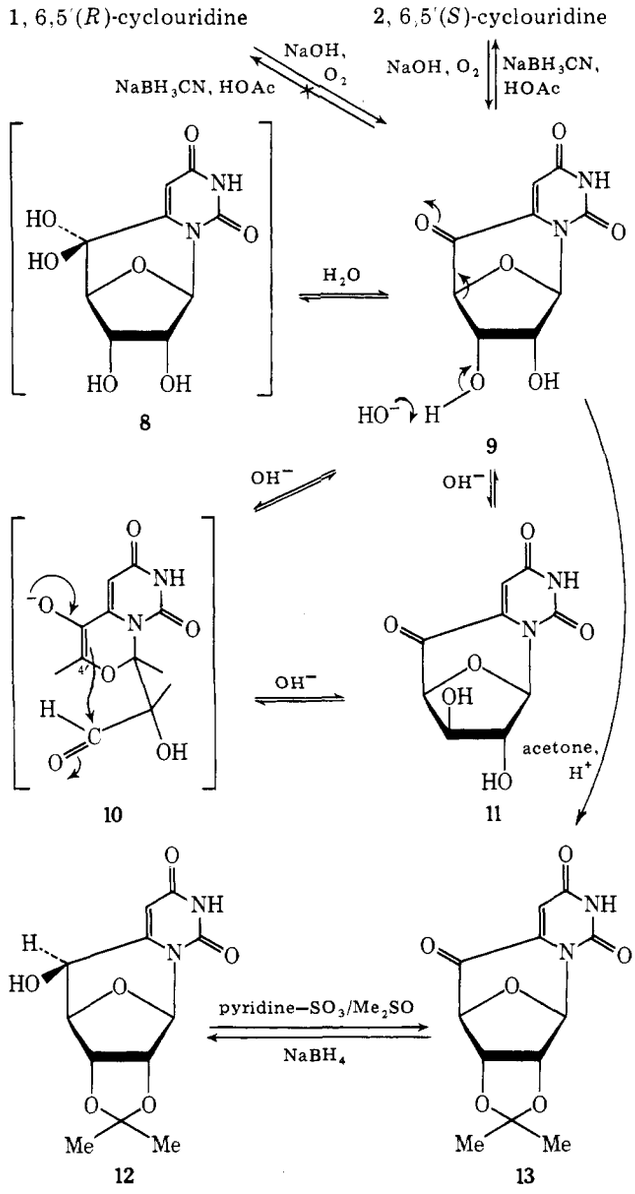
Scheme I



Scheme II



mediates required for the preparation of the 6,5′-cyclocytidine[5] analogues of 1 and 2. Thus, although methylation of thione 5 with diazomethane affords the expected 4-methylthio nucleoside 6, methylation of 5 or its tri-*O*-acetate 4 with methyl iodide in aqueous methanol at pH 9 affords, unexpectedly, the 5′-oxo-xylosyl nucleoside 7. The same product (7) is obtained when 6 is treated with aqueous sodium hydroxide in methanol (pH 9), and chromatography indicates that 6 is probably an intermediate in the conversion of 5 into 7. The structure of 7 and the manner of its formation were deduced from subsequent experiments with the 6,5′-cyclouridines 1 and 2 described below.

6,5′(*S*)-Cyclouridine (2) is stable in 1 N NaOH under nitrogen, but it is converted readily into the 5′-oxo-ribo nucleoside 9 in the presence of air or oxygen (Scheme II). Autoxidation of 6,5′(*R*)-cyclouridine (1) also affords 9, indicating that oxidation occurs at the 5′ position. Interestingly, the rate of oxidation of 1 is much slower than that of 2. That 9 retains the ribo configuration is evident from the NMR spectrum and from the fact that 9 can be converted into an isopropylidene derivative 13, identical with that obtained by sulfur trioxide–pyridine oxidation[6] of 12.

The keto nucleoside 9 is stable in 1 N NaOH, in which it is formed from 1 or 2, but under less strongly alkaline conditions it equilibrates with its 5′-oxo-xylo isomer 11. This isomerization is very rapid at pH 8–9, where the equilibrium favors the xylo nucleoside 11, and occurs at an appreciable rate simply on dissolving 9 in water. Preparatively, treatment of 9 with dilute ethanolic ammonia, followed by removal of ammonia by evaporation, affords a mixture from which the major isomer (11) crystallizes readily. Compound 11 correspondingly reequilibrates with its ribo isomer 9 on dissolution in water or dilute alkali. NMR studies show that the equilibrium 11 ⇌ 9 in 1 N NaOD lies entirely in favor of the ribo isomer 9, a finding that explains the apparent stability of 9 in 1 N NaOH.

The sequence of events occurring in the conversion of 4 into 7 (Scheme I) is therefore S-methylation to give 6, followed by autoxidation to give the 5′-oxo analogue of 6, which equilibrates with the observed xylo product 7 under the mild conditions used (pH 9).

The assignment of the xylo configuration to 7 and 11 rests on their NMR (Me_2SO-d_6), which show $J_{3',4'}$ values of 7.4 Hz and very small values (<0.5 Hz) for $J_{2',3'}$ (Table I). In contrast, the ribo epimer 9 shows $J_{3',4'} = 0$ and $J_{2',3'} = 6.3$ Hz. Similar differences were observed in the NMR spectra of the phenylhydrazones of 9 and 11. Additionally, both 7 and 11 show long-range coupling (1.2 Hz) between H-1′ and H-3′, which is consistent with the geometry of the xylo configuration. Four-bond couplings of similar magnitude have been observed previously for a variety of bicyclic carbohydrates.[7]

The most likely mechanism for the interconversion of 9 and 11 is a retro-aldol cleavage to generate the pyrimido[1,6-*c*]-

Table I. First-Order Coupling Constants,[a] Hz

Compd	$J_{2',3'}$	$J_{3',4'}$	$J_{4',5'}$	$J_{1',3'}$	$J_{5,5'}$	$J_{5,NH}$	$J_{5',5'OH}$	$J_{3',3'OH}$	$J_{2',2'OH}$
3	6.1	0	6.4	0	1.3	1.3			
4	6.2	0	6.2	0	1.3	1.3			
5	6.1	0	6.1	0	1.2	1.2	6.1	7.0	5.5
6	6.1	0	6.1	0	1.2		6.1	7.0	5.5
9[b,c]	6.3	0		0		2.1		*d*	*d*
9 (8)	6.1	0		0		ex	ex	ex	ex
9X[l]	6.4	0		0		1.8		6.4	5.5
7[e]	~0.5[f]	7.4		1.2				4.5	4.3
11[b]	~0.5[f]	7.4		1.2		*g*		4.4	4.3
11X[l]	1.2	6.7		1.2		1.1		*k*	5.2
16 (NaOD)[e,i]	~0.5[f]	6.6	5.9	1.2	1.2	ex	ex	ex	ex
14	~1.0[f]	7.3	~0.5	1.2[j]	~0.5	*k*	6.7	4.0	4.9
15[b]	0	6.4	6.1	1.1	1.2	2.0			4.3

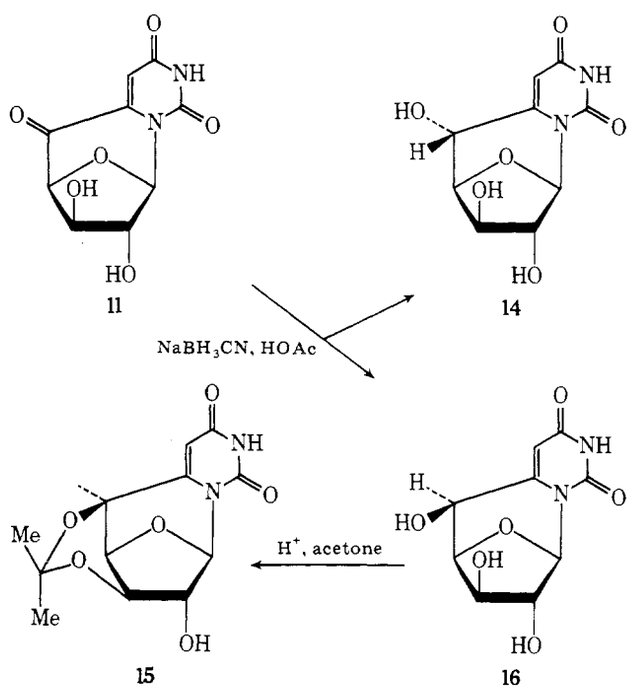
[a] In all cases $J_{1',2'} = 0$ Hz. Values for $J_{2',3'}$, $J_{3',4'}$, and $J_{4',5'}$ for compounds with unsubstituted hydroxyl groups were obtained after addition of D_2O. Computer resolution = 0.3 Hz unless stated otherwise. [b] Computer resolution = 0.15 Hz. [c] $^4J_{1',4'} = 0.76 \pm 0.15$ Hz. [d] First-order values not obtainable. [e] Computer resolution = 0.19 Hz. [f] $J_{2',3'}$ not resolved but detectable by decoupling. [g] Obscured by 2′-OH signal but detectable by decoupling. [h] Obscured by H-1′ and H-4′ signals. [i] $^4J_{3',5'} = 1.2 \pm 0.19$ Hz. The only first-order values obtainable for **16** in Me_2SO-d_6 are $J_{2',2'OH} = 4.5$ Hz and $J_{3',3'OH} = 4.0$ Hz. [j] Obtained after D_2O addition and decoupling $J_{2',3'}$. [k] Obscured by H-1′ signal. [l] Phenylhydrazone derivative.

[1,3]oxazine intermediate **10**, which can then recyclize to give either **9** or **11** depending on the orientation of the aldehyde group in the transition state. The rate of ring closure apparently exceeds the rate at which enolate **10** ketonizes because NMR studies of the **9** ⇌ **11** interconversion under a variety of alkaline conditions in D_2O show that deuterium is not incorporated at C-4′ of either **9** or **11**. Lack of deuterium incorporation, however, rules out the possibility that the 3′ epimerization involves abstraction of H-4′ and elimination of the 3′-hydroxyl group, followed by rehydration. This dehydration–rehydration sequence is in any case unlikely because formation of the olefinic intermediate would violate Bredt's rule. Further support for the retro-aldol mechanism comes from the fact that the isopropylidene nucleoside **13**, in which the 3′-hydroxy group is blocked, is stable to conditions that promote rapid equilibration of **9** and **11**.

A C-3′ epimerization reaction similar to that described above was observed recently by Youssefyeh et al.[8] during the base-catalyzed aldol coupling of formaldehyde with unprotected uridine 5′-aldehyde. Their reaction involves hydroxymethylation at C-4′ of uridine 5′-aldehyde, followed by Cannizzaro reduction of the original 5′-aldehyde group to give 4′-hydroxymethyluridine, together with its 3′ epimer.[9] These authors[8] also suggested a retro-aldol–aldol cyclization mechanism, and our results with the nonenolizable, constrained ketones **9** and **11** tend to support this proposal.

A curious feature of the retro-aldol equilibration of **9** and **11** is that the reaction itself, and the purity of individual preparations of **9** and **11**, can be monitored by UV spectroscopy. This follows from the finding that ribonucleoside **9** is largely hydrated in water and has a UV spectrum different from that of xylo nucleoside **11**, which exists in water in the keto form. In water, compound **9** absorbs strongly at 270 nm but shows a much smaller peak at 315 nm. In contrast, nucleoside **11** absorbs strongly at 312 nm and has no discrete peak at 270 nm. Removal of the 5′-carbonyl conjugation by hydration would be expected to result in a hypsochromic shift, and the 270-nm absorption of **9** can therefore be attributed to the hydrate **8**. In support of this conclusion, it should be noted that solutions of **9** in anhydrous dioxane absorb *only* at 321.5 nm but that addition of water to the dioxane solution results in the reappearance of absorption at 270 nm.[10] Further evidence for the existence of **8** comes from NMR studies of **9** in aqueous systems. Thus, the NMR spectrum of **9** in Me_2SO-d_6 consists of a single set of peaks (Table II), but addition of D_2O results in the gradual appearance of an additional set of peaks, attributable to **8**, which reach a constant 8/9 ratio of ~1:1. Similarly, the NMR spectrum of **9** in D_2O alone shows *two* sets of peaks, with a **8**/**9** ratio of 3.5:1. On the other hand, xylo nucleoside **11** gives a *single* set of peaks in D_2O and Me_2SO-d_6–D_2O that is closely similar to the spectrum in anhydrous Me_2SO-d_6, indicating in this case that the equilibrium lies heavily in favor of the keto form of **11**.

Scheme III



A further instance where the ribo (**9**) and xylo (**11**) 5′-keto nucleosides show disparate properties concerns their reduction with sodium cyanoborohydride in acetic acid. This reagent combination[11] (pH ~4) was used because alkaline solutions of sodium borohydride induce C-3′ epimerization, with consequent formation of mixtures of ribo- and xylo-6,5′-cyclopyrimidine nucleosides. Cyanoborohydride reduction of **9** gives 6,5′(*S*)-cyclouridine (**2**), with no detectable formation of the 5′*R* isomer **1**. Similar reduction of **11**, however, affords both the 5′*R* and 5′*S* isomers of 6,5′-cyclo-1-(β-D-xylofuranosyl)uracil (**14** and **16**, respectively, Scheme III) in a ratio of 1:5. Clearly, attack by the cyanoborohydride ion on **9** and **11** occurs in both cases primarily from the less hindered, rear side

Table II. Proton Chemical Shifts (δ) at 100 MHz[a]

Compd	Registry no.	N_3H	C_5H	$C_1'H$	$C_2'H$[b]	$C_3'H$[b]	$C_4'H$	$C_5'H$	O_5H	$O_2'H$[b]	$O_3'H$[b]	Other
3	64200-89-1	11.41	6.06 n m	5.74	5.41 d	5.60 d	4.72 d	5.82 dd				OAc 2.02, 2.12, 2.15
4	64200-88-0	12.76	6.39 t	5.76	5.44 d	5.61 d	4.74 d	5.76 dd				OAc 2.02, 2.12, 2.15
5	64200-87-9	12.63	6.39 n m	5.73	4.11 t	4.35 t	4.26 d	4.64 dt	6.51 d	5.40 d	5.26 d	
6	64200-86-8		6.51 d	5.78	3.99 t	4.35 t	4.28 d	4.75 dt	6.51 d	5.48 d	5.22 d	SMe 2.44
9[c]	64234-75-9	11.74	6.12 d	5.93	——4.34 m——		4.67			——5.61 d——		
9 (8)[d]	64200-83-5	ex	5.88	5.74	3.99 d	~4.3 d	4.06			ex	ex	
9X[e]	64234-74-8	11.25	6.05 d	5.93	4.15 dd	4.30 t	5.62			5.33	5.68	PhNH 10.66, Ph (4 H, m) 7.30, (1 H, m) 6.95
13	64200-82-4	11.84	6.15 d	6.07	———	4.97	———					Ip 1.44, 1.29
7	64200-81-3		6.87	5.85 d	4.07 d	4.48 m	5.15 d			6.19 d	6.24 d	SMe 2.50
11	64200-80-2	11.79	6.08 d	5.81 d	4.08 d	4.46 m	5.09 d			6.09 d	6.22 d	
11X[f]	64200-79-9	11.29	6.15 d	~5.77 d	3.94 dd	4.35 m	~5.74 d			6.00 d	~5.77 d	PhNH 10.37, Ph (4 H, m) 7.29, (1 H, m) 6.95
16	64234-73-7	11.33	——5.63 m——		3.89 d	4.41 m	4.65 m		4.95 m	5.86 d	6.03 d	
16[g]		ex	5.85 d	5.77 d	4.08 d	4.51 m	4.67 dd	4.93 dt	ex	ex	ex	
14	64234-72-6	11.34	——5.61 m——		3.72 d	4.18 m	4.56 d	4.44 d	6.07 d	5.81 d	5.69 d	Ip 1.24, 1.38
15	64200-78-8	11.39	5.57 dd	5.76 d	4.04 d	4.46 dd	5.05 t	4.86 dd		6.08 d		

[a] Spectra were obtained on a JEOL PFT-100 spectrometer operating in the Fourier transform mode (EC-100 computer) with an internal field frequency lock. Values given for chemical shifts are first order and (except for the N_3H signals of **4** and **5**) were obtained at 1250-Hz width with 8K data points, giving a computer resolution of 0.3 Hz. Unless otherwise stated, Me_2SO-d_6 was used as a solvent with Me_4Si as an internal standard. Peaks are singlets unless designated d (doublet), m (multiplet) dd (doublet of doublets), t (double doublets where coincidence of inner lines gives appearance of triplet), dt (double triplet or ddd), n (Narrow), or ex (exchanged). [b] The assignments of $C_2'H$–$O_2'H$ and $C_3'H$–$O_3'H$ can be interchanged for the ribosyl compounds (first eight entries) since the observed $J_{1',2'} = J_{3',4'} = 0$ Hz precludes unambiguous assignment. [c] Addition of D_2O reveals $C_2'H$ and $C_3'H$ as an AB quartet at δ 4.30 and 4.41. [d] Shifts for hydrated **9** (**8**) in Me_2SO-d_6–D_2O. [e] Phenylhydrazone derivative of **9**. [f] Phenylhydrazone derivative of **11**. [g] In 1 N NaOD, DSS standard.

Assignments of the 5′ configurations to **14** and **16** follow from their respective $J_{4',5'}$ values of <0.5 and 5.9 Hz. These values are diagnostic because Dreiding models show a 4′,5′ dihedral angle of ~90° for **14** and 30° for **16**. Both **14** and **16** show $^4J_{1',3'}$ values of 1.2 Hz, consistent with the xylosyl configuration, and **16** shows an additional four-bond coupling (1.2 Hz) between $H_{3'}$ and $H_{5'}$ that is consistent only with the 5′*S* configuration. Chemical proof of the 5′*S* configuration follows from the finding that **16** readily forms a 3′,5′-*O*-isopropylidene derivative **15**, whereas **14** is inert to acetone and *p*-toluenesulfonic acid because isopropylidene ring formation is sterically impossible. Compound **15**, in which the 2′-hydroxyl group is conveniently unblocked, is expected to be a versatile intermediate in further studies involving transformations of the sugar rings of these 6,5′-cyclopyrimidine nucleosides. We also anticipate that the retro-aldol C-3′ epimerization reaction will be applicable to other cyclonucleosides, e.g., 8,5′-cyclopurine nucleosides, of interest as probes of conformational aspects of enzyme–substrate interactions.

Experimental Section

General Procedures. Melting points were determined on a Thomas-Hoover apparatus (capillary method) and are uncorrected. Ultraviolet spectra were measured on Cary Model 15 and Varian Superscan 3 spectrometers and infrared spectra (KBr disk) were obtained with a Perkin-Elmer Infracord. Thin-layer chromatography was performed on 1 × 3 in. microscope slides coated with silica gel GF_{254} (Merck) and preparative separations were effected on 20 × 20 cm, 1-mm silica gel GF plates (Analabs Inc.). Separated materials were detected with ultraviolet light and/or by spraying with sulfuric acid in ethanol (10% v/v) followed by charring. Evaporations were carried out in vacuo with bath temperatures kept below 45 °C. Microanalyses were performed by Spang Microanalytical Laboratory, Ann Arbor, Mich.

2′,3′,5′-Tri-*O*-acetyl-6,5′(*S*)-cyclouridine (3).[13] 2′,3′-*O*-Isopropylidene-6,5′(*S*)-cyclouridine[4] (**12**, 1.18 g, 4.2 mmol) was dissolved with stirring in 80% acetic acid (40 mL), and the solution was refluxed for 8 h, at which time TLC (EtOAc) showed complete absence of starting material. The solution was concentrated to dryness, and pyridine (2 × 25 mL) was added to and evaporated from the residue. The final crystalline residue, comprising mostly 6,5′(*S*)-cyclouridine (**2**) together with small amounts of partially acetylated material, was dissolved in a mixture of pyridine (25 mL) and acetic anhydride (5 mL, 53 mmol). After 2 h at room temperature (TLC, EtOAc), ethanol was added to hydrolize excess acetic anhydride, and the mixture was evaporated to dryness. A solution of the residue in chloroform was washed with cadmium chloride solution to remove traces of pyridine. The organic layer was filtered, washed with water, and dried over sodium sulfate. Removal of the solvent afforded a dry foam which crystallized readily from warm ethanol to give 1.2 g (78%, TLC pure) of **3**, mp 209–210 °C.

Anal. Calcd for $C_{15}H_{16}N_2O_9$ (mol wt 368.30): C, 48.92; H, 4.38; N, 7.61. Found: C, 48.78; H, 4.40; N, 7.68.

2′,3′,5′-Tri-*O*-acetyl-6,5′(*S*)-cyclo-4-thiouridine (4). Phosphorus pentasulfide (1.0 g, 4.5 mmol) was added to a solution of **3** (1.0 g, 2.7 mmol) in 40 mL of dioxane, and the mixture was refluxed for 2 h (TLC, EtOAc/petroleum ether (30–60 °C), 1:1). The cooled mixture was filtered, and the filtrate and washings were concentrated to dryness. The solid residue was partitioned between dichloromethane and water; the organic layer was washed with water, dried over sodium sulfate, and evaporated to dryness. A solution of the residue in ~30

Table III. UV Data for 5′-Oxo-6,5′-cyclouridine (9)

Solvent	λ_{max} (ϵ)	λ_{max} (ϵ)	λ_{min} (ϵ)
Dioxane	321.5 (5460)		~250–280 (1540)
80% dioxane	321.5 (3750)	270 (4210)	295.5 (2420)
20% dioxane	318.5 (1670)	270 (7125)	299.5 (1300)
0.1 N HCl	315 (1420)	270 (8950)	300 (1240), 233 (1860)

mL of hot ethanol deposited 960 mg (92%) of **4** (yellow needles, TLC pure): mp 194–196 °C; UV λ_{max} (H_2O) 334, 279, 252 nm, λ_{min} 289, 262 nm.

Anal. Calcd for $C_{15}H_{16}N_2O_8S$ (mol wt 384.36): C, 46.87; H, 4.20; N, 7.29. Found: C, 46.85; H, 4.22; N, 7.26.

6,5′(*S*)-Cyclo-4-thiouridine (5). Four 1-mL portions of 1 N NaOH (4 mmol) were added over a 10-min period to a suspension of **4** (1.26 g, 3.28 mmol) in 40 mL of methanol. The solution was kept at room temperature for 1.5 h, when TLC (EtOAc) indicated complete hydrolysis. The solution was deionized by passage through excess Dowex 50 (H^+, previously equilibrated with methanol). Crystallization of **5** commenced on concentration of the effluent and was completed by cooling: yield 600 mg (73%); mp 249–250 °C; UV λ_{max} (H_2O) 250, 335 nm, λ_{min} 260 nm; λ_{max} (pH 11) 320, inflection 274–286 nm, λ_{min} 258 nm.

Anal. Calcd for $C_9H_{10}N_2O_5S$ (mol wt 258.25): C, 41.85; H, 3.90; N, 10.85. Found: C, 41.91; H, 3.94; N, 10.90.

6,5′(*S*)-Cyclo-4-methylthiouridine (6). A solution of **5** (400 mg, 1.55 mmol) in hot methanol (75 mL) was cooled rapidly to prevent crystallization. An excess of diazomethane in ether (dried over KOH) was added, and the solution was stored at room temperature until TLC (EtOAc) indicated that the reaction was complete. Removal of the ether and cooling afforded 200 mg (48%) of **6** with good TLC purity. Further crops contained traces of a faster moving component which was not fully characterized but which has an NMR spectrum consistent with the isomeric *N*-methyl compound (N–Me δ 3.56, Me_2SO-d_6). Recrystallization of **6** from methanol afforded pale yellow needles: mp 198–203 °C dec; UV λ_{max} (H_2O) 307, 225–238 (sh), 256–284 (sh), 313–320 nm (sh), λ_{min} 242 nm.

Anal. Calcd for $C_{10}H_{12}N_2O_5S$ (mol wt 272.28): C, 44.11; H, 4.44; N, 10.29. Found: C, 43.93; H, 4.50; N, 10.31.

6,5′-Cyclo-5′-oxo-1-(β-D-xylofuranosyl)-4-methylthiouracil (7). Method A. Methyl iodide (0.2 mL, 3.2 mmol) was added to a solution of **5** (100 mg, 0.39 mmol) in 15 mL of methanol, and the pH was adjusted to and maintained at ~9 by the dropwise addition of 1 N NaOH. TLC (CH_2Cl_2/MeOH, 9:1) at 30 min indicated the disappearance of starting material and the formation of **6**, which in turn was converted in to a faster moving component. After 3 h, the reaction mixture was neutralized with acetic acid, the volume was reduced, and the solution was applied to a preparative TLC plate. The plate was developed in CH_2Cl_2/MeOH (9:1), and the major zone was removed and extracted with 50 mL of EtOAc/MeOH (1:1). Concentration of the filtrate afforded pale yellow crystals of **7**: 60 mg (57%); mp 250–253 °C dec, darkens from 244 °C; UV λ_{max} (H_2O) 341, 229, 318–333 nm (sh), λ_{min} 265 nm; IR 1740 cm^{-1} (5′-oxo).

Anal. Calcd for $C_{10}H_{10}N_2O_5S$ (mol wt 270.26): C, 44.44; H, 3.72; N, 10.37. Found: C, 44.21; H, 3.76; N, 10.40.

Method B. A solution of **6** (20 mg) in methanol (1.5 mL) containing ~2 drops of 1 N NaOH was kept at room temperature for 3 h. Isolation of the product by preparative TLC as described above afforded 16 mg (81%) of material, identical (melting point, IR, UV, and NMR) with **7** prepared according to method A.

5′-Oxo-6,5′-cyclouridine (9). Method A. A slow stream of oxygen was passed through a solution of 6,5′(*S*)-cyclouridine (**2**, 500 mg) in 1 N NaOH (25 mL) for 60 h. The solution was neutralized by passage through excess Dowex 50 (H^+), and the effluent and washings (pH ~5) were concentrated to a clear syrup. Crystallization from ethanol afforded two crops of **9** (228 and 79 mg, total yield 62%). The mother liquors contained more **9**, together with xylo isomer **11**. An analytical sample of **9** was obtained by recrystallization from ethanol: mp 220–222 °C, resolidified, ~260 °C[14] dec; IR 1750 cm^{-1} (5′-oxo) (UV, see Table III).

Anal. Calcd for $C_9H_8N_2O_6$ (mol wt 240.17): C, 45.01; H, 3.38; N, 11.66. Found: C, 45.10; H, 3.40; N, 11.78.

Compound **9** readily forms a phenylhydrazone in methanol, mp >300 °C (recrystallized from 10% aqueous EtOH).

Anal. Calcd for $C_{15}H_{14}N_4O_5$ (mol wt 330.30): C, 54.55; H, 4.27; N, 16.96. Found: C, 54.38; H, 4.42; N, 16.74.

Method B. A solution of 6,5′(*S*)-cyclouridine[4] (**2**, 5 mg) in 0.5 mL of 1 N NaOD containing a trace of DSS was oxygenated in an NMR tube at room temperature. The NMR spectrum after 24 h showed complete conversion into **9** (hydrate), which shows signals at δ 5.97 (1, s, H-5), 5.83 (1, s, H-1′), 4.19 (1, d, H-3′, $J_{2',3'}$ = 6.1 Hz), and 3.98 (2, H-2′ d overlapping H-4′ s). This spectrum is identical with that of crystalline **9** in 1 N NaOD.

The above experiment was repeated on the same scale and under identical conditions with 6,5′(*R*)-cyclouridine[4] (**1**). The oxidation was considerably slower; after 92 h, integration indicated a **1**/**9** (hydrate) ratio of 3.2:1, with the signals for **9** (hydrate) (H-3′ obscured by H-4′ and H-5′ of **1**) identical with those above.

6,5′-Cyclo-5′-oxo-1-(β-D-xylofuranosyl)uracil (11). Method A. An aqueous ammonia solution (1 mL, 1 N) was added to a solution of **9** (80 mg) in warm ethanol (10 mL). The volume was reduced to 2 mL, and the solution was refrigerated, affording 47 mg (59%) of **11** (prisms): mp 260 °C dec, darkens above 240 °C; IR 1750 cm^{-1} (5′-oxo); UV λ_{max} (0.2 N HCl) 312 nm (ϵ 6200), λ_{min} 250 nm (ϵ 1485); λ_{max} (dioxane) 315 nm, λ_{min} 262 nm.

Anal. Calcd for $C_9H_8N_2O_6$ (mol wt 240.17): C, 45.01; H, 3.38; N, 11.66. Found: C, 44.88; H, 3.38; N, 11.64.

The mother liquors contained both **9** and **11**. Further crops of **11** can be obtained by treating the residue with ethanolic ammonia as above, followed by concentration and cooling of the solution.

Method B. 6,5′(*S*)-Cyclouridine[4] (**2**, 500 mg) was oxidized in 1 N NaOH as described above (method A) for the preparation of **9**. The clear syrup, obtained after evaporation of the deionized reaction mixture, was dissolved in 50 mL of ethanol. A 5-mL amount of a 1 N ammonia solution was added, and the solution was concentrated to ~10 mL and cooled. Crystalline **11** (279 mg) was collected; additional crops of 51 and 45 mg (total yield 75%) were obtained by retreating the residues with ethanolic ammonia. Compound **11** reisomerizes in 1 N NaOD to give ribonucleoside **9**, as shown by the change of the NMR spectrum [**11** (D_2O) δ 6.49 (s, H-5), 6.09 (d, H-1′, $J_{1',2'}$ = 1.2 Hz), 5.23 (d, H-4′, $J_{3',4'}$ = 7.6 Hz), 4.65 (dd, H-3′), 4.38 (s, H-2′)] to that described above for **9** (hydrate), preparation B.

Compound **11** forms a crystalline phenylhydrazone in 50% acetic acid, mp 275–280 °C dec, darkens above 265 °C (recrystallized from H_2O).

Anal. Calcd for $C_{15}H_{14}N_4O_5 \cdot H_2O$: C, 51.72; H, 4.63; N, 16.09. Found: C, 52.13; H, 4.28; N, 16.15.

2′,3′-*O*-Isopropylidene-5′-oxo-6,5′-cyclouridine (13). Method A. A suspension of **9** (55 mg) in acetone (3 mL) containing *p*-toluenesulfonic acid hydrate (15 mg) and 2,2-dimethoxypropane (0.1 mL) was stirred rapidly at room temperature. Further additions of dimethoxypropane (0.1 mL) were made after 1 and 3 h; TLC (EtOAc) indicated an essentially complete reaction after 4 h. The reaction mixture was neutralized by the addition of a saturated sodium bicarbonate solution, and the volume was reduced to ~0.5 mL. Crystalline **13** (34 mg, 53%) formed on the addition of water. The analytical sample was recrystallized from EtOAc/petroleum ether (bp 30–60 °C): mp 225–226 °C; UV λ_{max} (H_2O) 268, 316 nm, 268/316 = 7.0, λ_{min} 236, 298 nm, λ_{max} (dioxane) 320 nm, λ_{min} 268 nm. 268/316 = 0.34.

Anal. Calcd for $C_{12}H_{12}N_2O_6$ (mol wt 280.24): C, 51.43; H, 4.31; N, 10.00. Found: C, 51.16; H, 4.35; N, 9.80.

Method B. A solution of the sulfur trioxide–pyridine complex (477 mg, 3 mmol) in Me_2SO (1 mL) was added to a solution of **12** (282 mg, 1 mmol) in Me_2SO (1 mL) containing triethylamine (1 mL, 7 mmol), and the mixture was stored at room temperature for 17 h. The solution was acidified with glacial acetic acid and evaporated to dryness (lyophilization). Water was added to the residue, and crystalline starting material (**12**, 40 mg; TLC; NMR) was removed. The filtrate, which contains **13** and **12** as the main components, was applied to a preparative TLC plate. Development in benzene/ethyl acetate (1:2), followed by extraction of the appropriate zone with EtOAc and concentration to dryness, afforded pure **13** (95 mg) with melting point and IR, UV, and NMR spectra identical with **13** prepared as above. No attempt was made to optimize the yield of **13**.

Reduction of **13** (20 mg, 0.08 mmol) in methanol (5 mL) containing sodium borohydride (0.7 mL of a 1 N aqueous solution) for 30 min afforded a solution containing **12**, together with some faster moving (TLC, EtOAc) fluorescent materials. The identity of **12**, purified by preparative TLC, was established by comparison of the NMR spectrum with that of authentic material.[4]

6,5′(*S*)-Cyclouridine (2). Sodium cyanoborohydride (10 mg, 0.16 mmol) was added to a solution of **9** (36 mg, 0.15 mmol) in a mixture of methanol (2 mL) and acetic acid (0.5 mL). The solution was stored at room temperature for 1 h (TLC, EtOAc, dinitrophenylhydrazine spray) and then concentrated to dryness. An aqueous solution of the residue was passed through excess Dowex 50 (H^+), the eluate was evaporated to dryness, and methanol was repeatedly evaporated from the crystalline residue. The NMR spectrum (Me_2SO-d_6) of the re-

sulting crystalline mass (35 mg, 96%) was identical with that of authentic **2**;[4] none of the 5′*R* isomer **1** was detected, even with the very high signal-to-noise ratio resulting from prolonged spectral accumulation.

6,5′(*S*)-Cyclo-1-(β-D-xylofuranosyl)uracil (16) and 6,5′(*R*)-Cyclo-1-(β-D-xylofuranosyl)uracil (14). Sodium cyanoborohydride (40 mg, 0.64 mmol) was added to a suspension of **11** (150 mg, 0.53 mmol) in a mixture of water, acetic acid, and methanol (1:1:1, 6 mL). The mixture was stirred and warmed briefly to ~40 °C to effect dissolution and then cooled to room temperature. The reduction was monitored by the disappearance of the 310-nm peak of **11** and the appearance of absorption at 268 nm, a process that was complete after ~3.5 h. The solution was deionized by passage through an excess of Dowex 50 (H^+), the eluate and washings were concentrated to dryness, and methanol was repeatedly evaporated from the residue. The NMR spectrum (Me_2SO-d_6) of the residue showed compounds **14**/**16** in a ratio of 1:5. Pure **14** (80 mg) was obtained by crystallization of the residue from hot 80% ethanol: mp 246–248 °C; UV λ_{max} (H_2O) 268 nm, λ_{min} 233 nm; λ_{max} (pH 9) 266 nm, λ_{min} 241 nm.

Anal. Calcd for $C_9H_{10}N_2O_6$ (mol wt 242.19): C, 44.63; H, 4.16; N, 11.57. Found: C, 44.56; H, 4.14; N, 11.47.

A further sample of **14** (30 mg) and pure **16** (10 mg) was obtained by fractionation of the residue by preparative TLC (CH_2Cl_2/MeOH, 8:1; triple development). Compound **16** crystallized from aqueous ethanol: mp 250–253 °C dec, darkens and shrinks above 230 °C; UV λ_{max} (H_2O) 271 nm, λ_{min} 237 nm; λ_{max} (pH 10) 270 nm, λ_{min} 245 nm.

Anal. Calcd for $C_9H_{10}N_2O_6$ (mol wt 242.19): C, 44.63; H, 4.16; N, 11.57. Found: C, 44.86; H, 4.14; N, 11.36.

6,5′(*S*)-Cyclo-3′,5′-*O*-isopropylidine-1-β-D-xylofuranosyl)-uracil (15). A suspension of **16** (54 mg, 0.22 mmol) in acetone (3 mL) containing 15 mg of *p*-toluenesulfonic acid monohydrate and 0.1 mL of 2,2-dimethoxypropane was stirred rapidly at room temperature. The slow dissolution of **16** (~1 h) was followed by the appearance of crystalline **15**. After 3 h, the crystals (27 mg) were removed and washed with cold acetone. The filtrate was diluted with 0.1 mL of water, solid sodium bicarbonate was added, and the mixture was filtered. The filtrate was evaporated to dryness, and a solution of the residue in methanol was applied to a preparative TLC plate. After development (EtOAc), the appropriate zone was removed, the silica was extracted with ethyl acetate, and the filtrate was concentrated to dryness. Crystallization from 90% acetone afforded 20 mg (total yield 75%) of **15**: mp 265–266 °C, UV λ_{max} (H_2O) 269.5 nm, λ_{min} 233 nm; λ_{max} (pH 10) 270 nm, λ_{min} 243 nm.

Anal. Calcd for $C_{12}H_{14}N_2O_6$ (mol wt 282.25): C, 51.07; H, 5.00; N, 9.93. Found: C, 51.24; H, 5.05; N, 9.89.

Registry No.—**1**, 59728-02-8; **2**, 59686-60-1; **12**, 59686-58-7; acetic acid, 64-19-7; diazomethane, 334-88-3; methyl iodide, 74-88-4; phenylhydrazide, 100-63-0; 2,2-dimethoxypropane, 77-76-9.

References and Notes

(1) This investigation was supported by funds from the American Cancer Society (Grant CH-38) and from the National Institute of Health, U.S. Public Health Service (Grant No. 17085, for NMR studies).
(2) This paper is the second in a series entitled Conformationally Restricted Analogues of Pyrimidine Nucleosides. For Part 1, see ref 4.
(3) Reviews that stress the relationship between nucleoside–nucleotide conformation and biological activity include the following: (a) D. C. Ward and E. Reich, *Annu. Rep. Med. Chem.*, 272 (1969); (b) W. Saenger, *Angew. Chem., Int. Ed. Engl.*, **12**, 591 (1973). For examples of the use of conformationally restricted nucleotides to investigate enzyme specificity see the following: (a) A. Hampton, P. J. Harper, and T. Sakai, *Biochemistry*, **11**, 4965 (1972); (b) A. Matsuda, M. Tezuka, and T. Ueda, *Nucleic Acids Res., Spec. Publ.*, **No. 2**, S 13 (1976); (c) J. Zemlicka, *J. Am. Chem. Soc.*, **97**, 5896 (1975).
(4) B. A. Otter, E. A. Falco, and J. J. Fox, *J. Org. Chem.*, **41**, 3133 (1976).
(5) The synthesis of 6,5′(*S*)- and 6,5′(*R*)-cyclocytidine will be reported in a subsequent paper.
(6) J. R. Parikh and W. von E. Doering, *J. Am. Chem. Soc.*, **89**, 5505 (1967).
(7) (a) G. Kotowycz and R. U. Lemieux, *Chem. Rev.*, **73**, 669 (1973); (b) M. Barfield and B. Chakrabarti, *ibid.*, **69**, 757 (1969).
(8) R. Youssefyeh, D. Tegg, J. P. H. Verheyden, G. H. Jones, and J. G. Moffatt, *Tetrahedron Lett.*, 435 (1977). See also D. L. Leland and M. P. Kotick, *Carbohydr. Res.*, **38**, C9 (1974).
(9) It is not clear whether the C-3′ epimerization precedes or follows hydroxymethylation at C-4′ because the stability of uridine 5′-aldehyde alone in base was not reported. In either case, four initial products are possible since C-4′ can also epimerize, but this number is reduced to two (C-3′ epimers) because the subsequent Cannizzaro reaction removes the C-4′ asymmetry.
(10) Carbonyl hydration, and its effect on the UV spectrum, may be a common feature of 6-acylpyrimidines. Thus orotaldehyde (uracil 6-carboxaldehyde) absorbs in water (pH 1) at 261 nm (ε 8200), but it also shows a small shoulder at 300 nm (ε 700). On the basis of the above results, the 261-nm peak represents hydrated orotaldehyde, and the 300-nm peak can be attributed to the anhydrous form. In dioxane, orotaldehyde absorbs only at 300 nm. The literature value [K.-Y. Zee-Cheng and C. C. Cheng, *J. Heterocycl. Chem.*, **4**, 163 (1967)] for orotaldehyde is λ_{max} (pH 1–7) 261 nm (ε 13 300), with no mention of 200-nm absorption. We could not reproduce the ε 13 300 value, but we feel that our figure of ε 8200 is more in line with the value reported by the same authors for thymine 6-carboxaldehyde (ε 7800). In MeSO-d_6, anhydrous orotaldehyde shows NMR signals at δ 9.56 (s, CHO) and 6.28 (dd, H-5, $J_{N_3H,5} = J_{N_1H,5} = 1.8$ Hz). In D_2O, the anhydrous form [δ 9.60 (s, CHO), δ 6.49 (s, H-5)] and the hydrated form [δ 5.92 (d, H-5, $J_{allylic} = 1$ Hz), δ 5.75 (d CH$(OD)_2$)] are present in a ratio of ~1:15.
(11) B. F. Borch, M. D. Bernstein, and H. D. Durst, *J. Am. Chem. Soc.*, **93**, 2897 (1971).
(12) H. O. House, "Modern Synthetic Reactions", 2nd ed, W. A. Benjamin, New York, N.Y., 1972, pp 58–59.
(13) All of the compounds described can be named as substituted 6,9-epoxypyrimido[1,6-*a*]azepines, but for ease of comparison with ordinary nucleosides we prefer the trivial 6,5′-cyclonucleoside designations used herein.
(14) The melting point depends on the rate of heating. Examination of the UV spectrum of the resolidified melt shows that partial rearrangement to xylo isomer **11** occurs. Similarly, *aged* solutions of **9** in Me_2SO-d_6–D_2O rearrange to **11** on heating. In both cases, the reaction is probably catalyzed by alkali leached from the glass.

A Serendipitous Synthesis of 1,2,5,6-Tetramethyl-3,4,7,8-tetramethylenetricyclo[3.3.0.0^{2,6}]octane

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The title compound (**5**) can be prepared by photosensitized dimerization of 1,2-dimethyl-3,4-dimethylenecyclobutene (**1**) to *anti*-1,2,5,6-tetramethyl-3,4,7,8-tetramethylenetricyclo[4.2.0.0^{2,6}]octane (**2**), followed by flow system pyrolysis of **2** at 380 °C. At lower temperatures an intermediate, 1,2,5,6-tetramethyl-3,4,7,8-tetramethylenecycloocta-1,5-diene (**3**), can be isolated. On direct or sensitized photolysis of **3**, **5** is also obtained. The photochemistry of **2** has been explored, and its fragmentation to **1** on direct irradiation is discussed. The photosensitized dimerization of **1** to **2** is also discussed and interpreted in favor of a frontier orbital model for predicting the products of such reactions.

As an intermediate in a proposed synthesis, we required 1,2:5,6-bis(ethano)cyclooctatetraene (**4**, R = H) or a simple derivative thereof. Attempts to convert 1,2:5,6-bis(ethano)-cycloocta-1,5-diene[2] to the tetraene proved fruitless, and so we investigated the route to **4**, R = CH_3, shown in Scheme I. Our synthesis began with the photochemical dimerization of

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