Nucleosides. LXXVIII. Synthesis of Some 6-Substituted Uracils and Uridines by the Wittig Reaction[1]

ROBERT S. KLEIN AND JACK J. FOX*

Division of Organic Chemistry, Sloan-Kettering Institute for Cancer Research, Sloan-Kettering Division of Graduate School of Medical Sciences, Cornell University, New York, New York 10021

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Orotaldehyde (**1**) was treated with various alkylidene phosphoranes to afford α,β-unsaturated 6-substituted uracils. 6-Chloromethyluracil (**7**) was converted to the 6-triphenylphosphonium salt (**8**) by reaction with triphenylphosphine. This salt gave 6-styryluracil (**2**) when treated with benzaldehyde. With orotaldehyde, **8** afforded 1,2-bis(6-uracilyl)ethene (**14**), which was converted to 1,2-bis(6-uracilyl)ethane (**15**). With formaldehyde, **8** yielded 6-vinyluracil (**9**) which was polymerized to poly(6-vinyluracil) (**13**). Bromination of **9** afforded the 5-bromo analog **11** exclusively. With sodium bisulfite, **9** was converted quantitatively to the sodium salt of 2-(6-uracilyl)ethanesulfonic acid (**12**). Synthesis of 6-methylcytidine from N^4-acetyl-6-methylcytosine by the $Hg(CN)_2$–CH_3NO_2 procedure was achieved and the nucleoside was converted *via* a bisulfite adduct to 6-methyluridine, which was subsequently oxidized to tri-*O*-acetylorotidine aldehyde (**21**). With carbethoxymethylenetriphenylphosphorane, **21** was converted to the ethyl ester of *trans*-3-(6-uridinyl)acrylic acid (**23**).

Pyrimidine nucleosides containing a carbon substituent at C-6 have been generally difficult to synthesize. Simple examples of the C-6 methylated members of that group have been described recently.[2] More complex 6-alkylated nucleosides have been synthesized[3] by Claisen-type rearrangements of certain 5-allyloxy or 5-propynyloxy pyrimidine nucleosides to afford carbon to carbon 6-substituted derivatives bearing an hydroxy or ether function on C-5. A more general approach to various C–C 6-substituted nucleosides would be from the hiterto unknown uridine-6-carboxaldehyde ("orotidine aldehyde") by use of the Wittig reaction. Indeed, such a reaction was reported in the special case of 2-amino-4-hydroxy-5-phenylbutylpyrimidine-6-carboxaldehyde and its 2-acetamido derivative by Baker and Jordaan.[4] We report herein results on the use of the Wittig reaction on orotaldehyde (uracil-6-carboxaldehyde) and orotidine aldehyde as part of our general program dealing with the synthesis of nucleosides of potential biological interest. We also report a facile synthesis of 6-vinyluracil and its polymerization to poly(6-vinyluracil). The latter should be useful as a model system (with a polymethylene-type backbone) for use in binding studies with certain synthetic or naturally occurring polynucleotides.[5]

Attempts to condense orotaldehyde (**1**)[6] with ylides not stabilized by conjugation were unsuccessful. Thus, only trace amounts of 6-vinyluracial were detected, after chromatography, when **1** (Scheme I) was treated with methyltriphenylphosphonium bromide in dimethyl sulfoxide containing sodium methylsulfinylmethide. Better results were obtained, however, when **1** reacted with the stabilized ylide derived from benzyltriphenylphosphonium chloride in the presence of a threefold excess of phenyllithium in *N*,*N*-dimethylformamide (DMF) or with sodium methoxide to afford *trans*-6-styryluracil (**2**) in 51% yield. Trans isomerism in **2** was established by its pmr spectrum, which showed a characteristic coupling constant of 16.5 Hz for the vinylic protons. The corresponding cis isomer was not detected in the condensation products. The success of the reaction indicated that in spite of the charge in the anionic species of **1**, the aldehyde was still susceptible to nucleophilic attack by the ylide. (It is reasonable to expect that most of **1** is ionized in such strongly basic medium.) Hydrogenation of **2** over palladium/charcoal afforded the known 6-phenethyluracil (**3**).[7] Reaction of **1** with an equimolar amount of carbethoxymethylenetriphenylphosphorane in DMF gave **4** as the trans isomer in 62% yield. A trace of the cis isomer (detected by tlc) was found in the reaction mixture. The pK_a of this ylide is ~ 9[8] (*i.e.*, its pK_a is comparable to that of **1**) and, under the reaction conditions employed, appreciable amounts of the neutral form of the reactants present would favor condensation to **4**. When DMF was replaced by ethanol as the solvent of reaction, the crystalline product isolated in 95% yield was found by pmr spectroscopy to consist of a 15:85 mixture of the cis and trans isomers of **4**. The identity of each component in the mixture was determined by the different coupling constants of the vinylic protons: $J_{trans} = 16.5$ and $J_{cis} = 13.0$ Hz. These results are consistent with previous studies on the influence of the reaction medium on the stereoselectivity of the Wittig reaction with stabilized ylides[9] which concluded that "the proportion of the cis isomer is enhanced by the presence in the reaction solution of a Lewis acid such as a proton-donating solvent..."

Attempts were made to prepare certain 5-substituted orotaldehydes by oxidation of the corresponding 6-methyluracils with selenium dioxide. With 5-acetamido-6-methyluracil, no reaction occurred, while with the 5-nitro analog many side products were observed along with only trace amounts of the 6-aldehyde derivative. With 5-bromo-6-methyluracil, selenium dioxide oxidation did take place without side reactions. How-

(1) This investigation was supported in part by funds from the National Cancer Institute, National Institutes of Health, U. S. Public Health Service (Grant No. CA 08748).

(2) (a) M. W. Winkley and R. K. Robins, *J. Org. Chem.*, **33**, 2822 (1968); (b) M. Prystaš and F. Šorm, *Collect. Czech. Chem. Commun.*, **34**, 2316 (1969).

(3) B. A. Otter, A. Taube, and J. J. Fox, *J. Org. Chem.*, **36**, 1251 (1971); B. A. Otter, S. S. Saluja, and J. J. Fox, *ibid.*, **37**, 2858 (1972).

(4) B. R. Baker and J. H. Jordaan, *J. Heterocycl. Chem.*, **3**, 315, 319, 324 (1966).

(5) The syntheses of 1-vinyluracil and poly(1-vinyluracil) have been reported[5a] as well as the binding properties of the latter with certain synthetic polynucleotides.[5b] (a) J. Pitha and P. O. P. Ts'O, *J. Org. Chem.*, **33**, 1341 (1968); J. Pitha, P. M. Pitha, and P. O. P. Ts'O, *Biochim. Biophys. Acta*, **204**, 39 (1970); (b) J. Pitha, P. M. Pitha, and E. Stuart, *Biochemistry*, **10**, 4595 (1971).

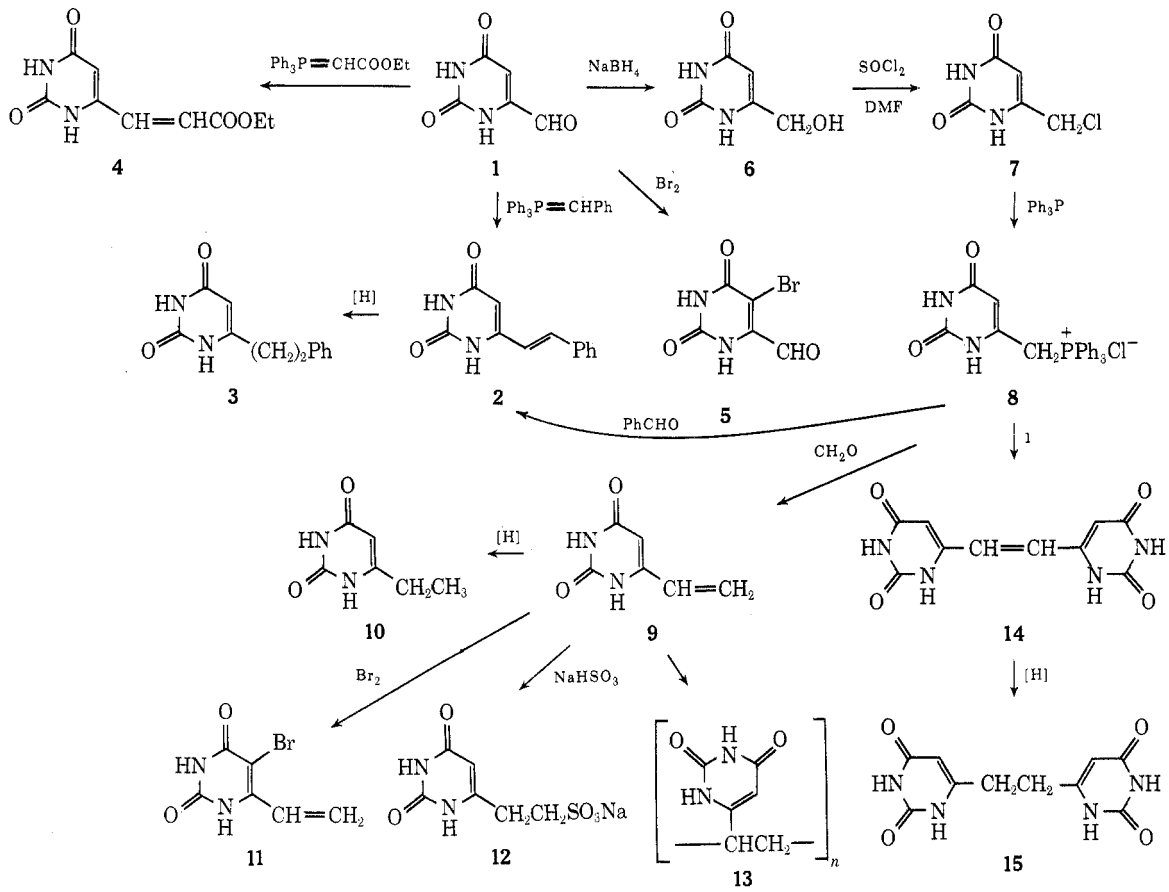
(6) (a) K.-Y. Zee-Cheng and C. C. Cheng, *J. Heterocycl. Chem.*, **4**, 163 (1967); (b) I. A. Mikhailopulo, V. I. Gunar, and S. I. Zav'yalov, *Izv. Akad. Nauk SSSR, Ser. Khim.*, 1388 (1967); *Chem. Abstr.*, **68**, 59523e (1968).

(7) B. R. Baker and M. Kawazu, *J. Med. Chem.*, **10**, 311 (1967).

(8) S. Fliszar, R. F. Hudson, and G. Salvadori, *Helv. Chim. Acta*, **46**, 1580 (1963); A. J. Speziale and K. W. Ratts, *J. Amer. Chem. Soc.*, **85**, 2790 (1963).

(9) H. O. House, V. K. Jones, and G. A. Frank, *J. Org. Chem.*, **29**, 3327 (1964).

SCHEME I



ever, the formation of product was very slow. A more practical approach to 5-bromoorotaldehyde (**5**) was found in the direct bromination of **1** with 1 equiv of bromine in water.

An alternate route to α,β-unsaturated 6-substituted uracils was found by condensation of aldehydes with the pyrimidyl Wittig derivative (**8**) which was prepared conveniently from **1** *via* **6** and **7**. Reaction of **8** with benzaldehyde gave, as expected, the styryl derivative **2**. When treated under nitrogen with formaldehyde and then with a threefold molar excess of sodium ethoxide in ethanol, compound **8** afforded 6-vinyluracil (**9**) in good yield. An excess of base is required for this reaction because of prior ionization of an NH proton in **8** before formation of the ylide intermediate. Hydrogenation of **9** over palladium/charcoal afforded the known[10] 6-ethyluracil (**10**) which, in addition to pmr data, confirms the structure of **9**.

The chemical properties of 6-vinyluracil are of interest. Bromination of **9** with an equivalent amount of bromine in water yielded exclusively 5-bromo-6-vinyluracil (**11**). When treated with bisulfite, compound **9** was converted quantitatively into the sulfonate salt **12**. This addition, unlike that of uracil, is not reversible in alkali.[11] The structure of **12** was easily established by the uv spectrum, which is similar to that for 6-ethyluracil, and the pmr spectrum, which exhibits signals for four methylenic protons and the C-5 vinylic proton. Attempts to achieve a free-radical polymerization of **9** were precluded by its poor solubility in most organic solvents. It was possible, however, to prepare the silylated derivative of **9**, which polymerized directly in dioxane solution and in the presence of azobisisobutyronitrile as free-radical initiator to silylated poly(6-vinyluracil) which, after hydrolysis, afforded poly(6-vinyluracil) (**13**) as a sparingly soluble, partially hydrated amorphous solid. The uv spectrum of this polymer is similar, as expected, to that for 6-alkyluracils.

The phosphonium salt (**8**) reacted with orotaldehyde (**1**) in DMF in the presence of excess sodium ethoxide to afford the diuracilyl derivative (**14**) in good yield. The elemental analyses and uv properties are consistent with structure **14** although, owing to the poor solubility of this compound, pmr measurements could not be made to determine its geometrical isomerism. Hydrogenation of **14** in aqueous base over palladium/charcoal gave the expected 1,2-bis(6-uracilyl)ethane (**15**), which also exhibited poor solubility properties. [It should be noted that **15** is structurally similar to 1,2-bis(3,4-dioxopiperazin-1-yl)ethane, a compound which has demonstrated[12] antitumor activity.]

In order to apply the Wittig reaction to nucleosides, a synthesis of orotidine aldehyde (**21**) (Scheme II) was achieved from N^4-acetyl-6-methylcytosine. Condensation of the latter with tri-*O*-benzoyl-D-ribofuranosyl bromide by the $Hg(CN)_2$–nitromethane procedure[13]

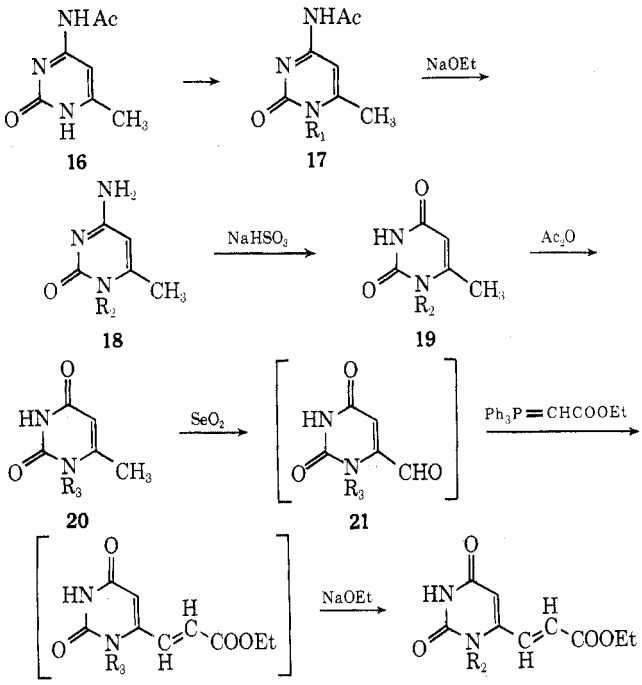
(10) (a) R. M. Evans, P. G. Jones, P. J. Palmer, and F. F. Stephens, *J. Chem. Soc.*, 4106 (1956); (b) R. Robinson and M. L. Tomlinson, *ibid.*, 1283 (1935).

(11) (a) R. Shapiro, R. E. Servis, and M. Welcher, *J. Amer. Chem. Soc.*, **92**, 422 (1970); (b) H. Hayatsu, Y. Wataya, K. Kai, and S. Iida, *Biochemistry*, **9**, 2858 (1970).

(12) A. M. Creighton, K. Hellmann, and S. Whitecross, *Nature (London)*, **222**, 384 (1969).

(13) (a) N. Yamaoka, K. Aso, and K. Matsuda, *J. Org. Chem.*, **30**, 149 (1965); (b) K. A. Watanabe and J. J. Fox, *J. Heterocycl. Chem.*, **6**, 109 (1969).

Scheme II



gave the blocked N-1 ribosylated pyrimidine (**17**) which, without isolation, was deacylated to 6-methylcytidine (**18**) in good yield.[14] Formation of 6-methyluridine (**19**) from **18** was performed using a method developed for the deamination of cytidine by catalysis with bisulfite[11a] under more stringent conditions. The relatively slow rate observed for this reaction as compared with that for cytidine is presumed to be due to the steric hindrance at C-6 by the 6-methyl group. After acetylation of **19** the protected nucleoside **20** was oxidized with selenium dioxide in 11:1 dioxane–glacial acetic acid to the tri-*O*-acetylorotidine aldehyde **21**. If the oxidation was carried out in glacial acetic acid, glycosyl cleavage of the product **21** occurred with the formation of orotaldehyde (**1**), while omission of HOAc slowed the oxidation with selenium dioxide considerably. Aldehyde **21** was unstable and, even in chloroform, it decomposed on standing. A small sample of **21** was purified by preparative tlc to a syrup which exhibited the expected pmr singals in $CHCl_3$ (δ 9.53 for aldehydic proton). The crude product **21** was treated immediately with carbethoxymethylenetriphenylphosphorane in DMF to afford the α,β-unsaturated ester derivative **22** which, after treatment with sodium ethoxide, gave crystalline *trans*-3-(6-uridinyl)acrylic acid as the ethyl ester **23**. Proof of **23** as the trans isomer is based on its pmr spectrum, which exhibited a coupling constant for the vinylic protons of the side chain of 16.0 Hz.

Extension of this approach to the synthesis of variously 6-substituted pyrimidine nucleosides is now underway in our laboratories.

(14) Syntheses of 6-methylcytidine and 6-methyluridine have been reported[2] by other methods. However, the reported yields were lower and the isolation of product was more laborious.

Experimental Section

Melting points were obtained on a Thomas-Hoover apparatus (capillary method) for temperatures below 300° and are uncorrected. For higher temperatures, a Mel-Temp block was used. The pmr spectra were measured on a Varian A-60 spectrometer using TMS as internal standard. Chemical shifts are reported in parts per million (δ). Signals are described as s (singlet), d (doublet), t (triplet), q (quartet), or m (complex multiplet). Values given for coupling constants and chemical shifts are first order. Uv spectra were measured on a Unicam SP 800-B ultraviolet spectrophotometer. Thin layer chromatography was performed on silica gel GF_{254} (Merck); spots were detected by uv absorbance or by charring after spraying with 20% v/v sulfuric acid–ethanol. Microanalyses were performed by Spang Microanalytical Laboratory, Ann Arbor, Mich., and by Galbraith Laboratories, Knoxville, Tenn. All evaporations were carried out *in vacuo*.

6-Styryluracil (2). A. From Orotaldehyde (1).—A mixture of anhydrous orotaldehyde[6a] (0.70 g, 0.0050 mol) and triphenylbenzylphosphonium chloride (1.94 g, 0.0050 mol) was dissolved in 25 ml of dry DMF under a slow stream of nitrogen. To the solution was added 7.5 ml of a 2 *M* solution (~0.015 mol) of phenyllithium in benzene–ether (70:30). The mixture was heated for 30 min at ~90° and cooled to ambient temperature. The solution was neutralized with glacial acetic acid and evaporated to dryness. The crystalline residue was recrystallized from ethanol to afford 0.550 g (51%) of the product in two crops. One final recrystallization from glacial acetic acid gave an analytically pure sample of **2**: mp 345–350° dec; pmr (DMSO-d_6) δ 5.77 (1, s, H-5), 6.84 (1, d, $CH{=}CHC_6H_5$), 7.30–7.90 (6, m, CHC_6H_5), 10.83 (2, broad singlet, 2 NH's), J_{trans} = 16.5 Hz.

Anal. Calcd for $C_{12}H_{10}N_2O_2$: C, 67.28; H, 4.71; N, 13.08. Found: C, 67.13; H, 4.76; N, 13.11.

B. From 8.—To a solution of **8** (*vide infra*) (2.12 g, 0.0045 mol) and 0.530 g (0.005 ml) of benzaldehyde in 100 ml of ethanol was added under a slow stream of dry nitrogen a solution of sodium ethoxide [from 0.350 g (0.0150 g-atom) of sodium metal] in 25 ml of ethanol. The mixture was heated to reflux for 15 min, cooled, and neutralized with glacial acetic acid. The crude product **2** (0.305 g, 31%) was collected by filtration and recrystallized from glacial acetic acid to give an analytically pure sample of **2** identical in all respects with the sample obtained by method A.

6-Phenethyluracil (3).—A solution of **2** (0.524 g, 0.00245 mol) in 200 ml of glacial acetic acid was hydrogenated at 1 atm over 20 mg of 10% Pd/C. After hydrogen uptake had ceased, the mixture was filtered through Celite and evaporated to dryness. The residue crystallized from glacial acetic acid in three crops to afford 0.461 g (87%) of product **3**. One more recrystallization from water–ethanol gave an analytical sample: mp 254–256° dec (lit.[7] mp 260–262°); pmr (DMSO-d_6) δ 2.30–2.90 (4, m, CH_2CH_2, overlapping DMSO), 5.33 (1, s, H-5), 7.25 (5, s, C_6H_5), 10.89 (2, broad singlet, 2 NH's).

Anal. Calcd for $C_{12}H_{12}N_2O_2$: C, 66.65; H, 5.59; N, 12.95. Found: C, 66.53; H, 5.71; N, 12.96.

***trans*- and *cis*-3-(6-Uracilyl)acrylic Acid Ethyl Ester (4).**—To a magnetically stirred suspension of anhydrous orotaldehyde (1.40 g, 0.0100 mol) in 30 ml of dry DMF was added 3.48 g (0.0100 mol) of carbethoxymethylenetriphenylphosphorane. The reaction was slightly exothermic. After 1.5 hr, ethanol (30 ml) was added to the clear solution, and the mixture was cooled at 0°. The crystalline product, collected by filtration, weighed 1.31 g (62%). One recrystallization from ethanol gave analytically pure material: mp 255–257°; uv $\lambda_{max}^{pH\ 1}$ 305 mμ (ϵ 10,900), $\lambda_{min}^{pH\ 1}$ 265 (3500), $\lambda_{max}^{pH\ 11}$ 336 (7900), $\lambda_{min}^{pH\ 11}$ 283 (1900); pmr (DMSO-d_6) δ 1.26 (3, t, CH_2CH_3), 4.22 (2, q, CH_2CH_3), 6.02 (1, s, H-5), 6.89 and 7.28 (2, two AB doublets, $CH{=}CH$), 10.95 and 11.15 (2, two broad singlets, 2 NH's), J_{trans} = 16.5 Hz.

Anal. Calcd for $C_9H_{10}N_2O_4$: C, 51.43; H, 4.80; N, 13.33. Found: C, 51.39; H, 4.76; N, 13.27.

The reaction was repeated on the same scale using 40 ml of ethanol instead of DMF as the solvent. The mixture was heated to reflux for 1.5 hr, chilled, and filtered. The crystalline product weighed 2.00 g (95%). Pmr measurements indicated that the product was a 15:85 mixture of the cis and trans isomers. No attempt was made to isolate each isomer from the mixture. Pmr signals corresponding to the cis isomer of the mixed spectrum are described as follows: pmr (DMSO-d_6) δ 1.09 (3, t,

CH_2CH_3), 4.23 (2, q, CH_2CH_3), 5.70 (1, s, H-5), 6.30 and 6.75 (2, two AB doublets, CH=CH), J_{cis} = 13.0 Hz.

When the reaction was performed in methanol, the final product (95% yield) consisted of a 1:4 mixture of the cis and trans isomers.

5-Bromorotaldehyde (5).—To a solution of orotaldehyde monohydrate (1.58 g, 0.0100 mol) in 500 ml of water was added dropwise a saturated solution of 1.70 g of bromine in water. After complete decolorization, the mixture was treated with sodium acetate trihydrate (1.36 g, 0.0100 mol), evaporated to a small volume, and chilled. The precipitate obtained in three crops was filtered and washed with cold water. The white crystalline product (5) weighed 1.85 g (78%). One recrystallization from water gave the analytical sample: mp 273–275° dec; uv $\lambda_{max}^{pH\ 1}$ 279 mμ (ε 7800), $\lambda_{min}^{pH\ 1}$ 241 (1400), $\lambda_{max}^{pH\ 11}$ 301 (6600), $\lambda_{min}^{pH\ 11}$ 253 (1600), $\lambda_{max}^{pH\ 14}$ 286 (6300), $\lambda_{min}^{pH\ 14}$ 253 (2400).

Anal. Calcd for $C_5H_3BrN_2O_3$: C, 27.42; H, 1.38; N, 12.79; Br, 36.49. Found: C, 27.32; H, 1.44; N, 12.84; Br, 36.47.

5-Bromorotaldehyde Phenylhydrazone.—As the pmr of 5 in DMSO-d_6 was complicated by what is presumed to be a dimeric structure, the phenylhydrazone derivative of 5 was prepared as a further confirmation of its structure: mp 264° dec; pmr (DMSO-d_6) δ 6.70–7.60 (5, m, C_6H_5), 7.93 (1, s, -CH=N), 11.21 (3, broad singlet, 3 NH's). When TFA was added the last signal resolved into two singlets at δ 10.79 (1 NH) and 11.45 (2 NH's).

Anal. Calcd for $C_{11}H_9BrN_4O_2$: C, 42.74; H, 2.93; N, 18.12; Br, 25.85. Found: C, 42.69; H, 2.89; N, 18.10; Br, 25.82.

6-Chloromethyluracil (7).—To a well-stirred suspension of very finely ground 6-hydroxymethyluracil monohydrate 6[15,16] (3.10 g, 0.0194 mol) in 20 ml of thionyl chloride was added 1 ml of $SOCl_2$ containing 8 drops of DMF. After an initial vigorous evolution of HCl gas the mixture was heated to reflux for 5–10 min. As the reaction proceeded the solids that precipitated on the flask walls were washed back into the mixture with 10 ml of thionyl chloride. It was found necessary occasionally to break big clumps as they formed. The mixture was then cooled to room temperature and diluted with 70 ml of diethyl ether and the crude product was collected by filtration and washed with ether (2.90 g, 93%). One recrystallization from hot acetic acid gave 1.45 g of pure material, mp 249–251° (lit.[17] mp 240° dec). A second crop, mp 244–248°, brought the total yield to 2.30 g (74%). This was used without further purification: pmr (DMSO-d_6) δ 4.40 (2, s, CH_2Cl), 5.70 (1, s, H-5), 11.11 (2, broad singlet, 2 NH's).

6-Uracilylmethylenetriphenylphosphonium Chloride (8).—A solution of 7 (3.60 g, 0.0225 mol) and triphenylphosphine (9.00 g, 0.0343 mol) in 250 ml of ethanol was heated to reflux for 16 hr. Another 3.2 g (0.0122 mol) of triphenylphosphine was then added and heating was resumed for another 3 hr while 100 ml of ethanol was removed by distillation. The clear solution was chilled and the crystalline product (7.25 g) was collected. Evaporation of the mother liquor afforded another crop to give a total yield of 9.00 g (84%). Two recrystallizations from ethanol gave the analytical material: mp 200° dec; uv $\lambda_{max}^{pH\ 1}$ 265.5 mμ (ε 8600), $\lambda_{min}^{pH\ 1}$ 246 (5400); pmr (DMSO-d_6) δ 1.05 (3, t, CH_3 of ethanol), 3.46 (2, q, CH_2 of ethanol), 5.18 (2, broad singlet, CH_2), 5.46 (1, s, H-5), 7.55–8.10 (15, m, 3 C_6H_5), 11.06 and 11.23 (2, 2 broad singlets, 2NH's). The presence of 1 mol of ethanol supports the analytical data.

Anal. Calcd for $C_{23}H_{20}N_2O_2PCl \cdot C_2H_5OH$: C, 64.03; H, 5.59; N, 5.97; P, 6.60; Cl, 7.56. Found: C, 63.32; H, 5.60; N, 5.98; P, 6.61; Cl, 7.84.[18]

6-Vinyluracil (9).—A mixture of the phosphonium chloride 8 (0.423 g, 0.0009 mol) and paraformaldehyde (0.135 g, 0.0045 mol) was suspended in 30 ml of absolute ethanol. To the stirred mixture, which was kept under nitrogen,[19] was slowly added ~0.003 mol of sodium ethoxide (from 75 mg of sodium metal) in 10 ml of ethanol. The mixture became yellow and then slowly decolorized as a precipitate formed. The suspension was then heated to 50° for 5 min, cooled to room temperature, neutralized with glacial acetic acid to pH 4–5, and filtered. After evaporation of the filtrate to a small volume some insoluble material that had precipitated was again removed and the clear solution was evaporated to dryness. The residue, which contains triphenylphosphine oxide in addition to 9, was partitioned between chloroform and water and the aqueous extract was treated with an excess of Dowex 50 W (H^+) resin. After filtration, the solution was evaporated to dryness and the crystalline residue was recrystallized from hot acetic acid to afford pure 9 in three crops (0.104 g, 83%). Another recrystallization from acetic acid gave an analytically pure sample: mp >350°; uv $\lambda_{max}^{pH\ 1}$ 296.5 mμ (ε 9200), $\lambda_{min}^{pH\ 1}$ 247 (1900), $\lambda_{max}^{pH\ 11}$ 307 (7000), $\lambda_{min}^{pH\ 11}$ 257 (1500), $\lambda_{max}^{pH\ 14}$ 308 (6300), $\lambda_{min}^{pH\ 14}$ 260 (1600); pmr (pyridine-d_6) quartet centered at δ 5.67 (1, RCH_α=CH_{trans} H_{cis}) quartet centered at 6.27 (1, H_{trans}), quartet centered at 6.61 (1, H_α), ~12.5 (2, broad singlet, 2 NH's), J_{gem} = 2.5–3.0 Hz, J_{trans} = 17.5–18.0 Hz, J_{cis} = 8.5–9.0 Hz.

Anal. Calcd for $C_6H_6N_2O_2$: C, 52.17; H, 4.38; N, 20.28. Found: C, 51.94; H, 4.33; N, 20.13.

6-Ethyluracil (10).—Compound 9 (0.276 g, 0.0020 mol) was dissolved in 100 ml of hot water and catalytically reduced under hydrogen (1 atm) in the presence of 10% Pd/C at ambient temperature. The uptake was rapid (16 min) and quantitative. After concentration, the filtrate deposited a first crystalline crop (0.198 g) of 10, mp 204–207° (lit.[10b] mp 205°). A second crop (0.045 g) was obtained after further evaporation to give a total yield of 87%: pmr (DMSO-d_6) δ 1.09 (3, t, CH_3), 2.31 (2, q, CH_2), 5.31 (1, s, H-5), 10.75 (2, broad singlet, 2 NH's).

5-Bromo-6-vinyluracil (11).—To a vigorously stirred solution of 6-vinyluracil (0.138 g, 0.0010 mol) in 50 ml of water was added, dropwise, an aqueous solution of bromine (0.176 g, 0.0011 mol/10 ml H_2O). After 30 min the mixture was chilled and the white crystalline product was collected by filtration (0.130 g), decomposes slowly over 250°. Another crop of 11 (0.157 g, total yield 72%) was obtained after evaporation of the mother liquor: uv $\lambda_{max}^{pH\ 1}$ 305 mμ (ε 8600), $\lambda_{min}^{pH\ 1}$ 255 (1100), $\lambda_{max}^{pH\ 11}$ 327 (7000), $\lambda_{min}^{pH\ 11}$ 268 (1000), $\lambda_{max}^{pH\ 14}$ 325 (5900), $\lambda_{min}^{pH\ 14}$ 272 (1200); pmr (DMSO-d_6) quartet centered at δ 5.85 (1, RCH_α=CH_{trans} H_{cis}), quartet centered at 6.32 (1, H_{trans}), quartet centered at 6.86 (1, H_α), 11.10 and 11.52 (2, 2 broad singlets, 2 NH's), J_{gem} = 0.5–1.0 Hz, J_{trans} = 17.5 Hz, J_{cis} = 11 Hz.

Anal. Calcd for $C_6H_5BrN_2O_2$: C, 33.21; H, 2.32; N, 12.91; Br, 36.82. Found: C, 33.08; H, 2.49; N, 12.93; Br, 36.85.

2-(6-Uracilyl)ethanesulfonic Acid (Sodium Salt) (12).—To a solution of 6-vinyluracil (0.138 g, 0.0010 mol) in 40 ml of water was added a solution of sodium bisulfite (0.125 g, 0.0012 mol) in 5 ml of water and the clear mixture was heated on the steam bath for 20 min. Uv measurements indicated that the $\lambda_{max}^{pH\ 1}$ at 307 mμ disappears as a new maximum at 261 mμ increases in intensity. Evaporation of the mixture and crystallization from water–methanol afforded 0.229 g (88%) of the crystalline sulfonate as the monohydrate in three crops: mp 318–321° dec with loss of water at 110°; uv $\lambda_{max}^{pH\ 1}$ 261 mμ (ε 8000), $\lambda_{min}^{pH\ 1}$ 229 (1500), $\lambda_{max}^{pH\ 11}$ 283 (7000), $\lambda_{min}^{pH\ 11}$ 242 (1400), $\lambda_{max}^{pH\ 14}$ 278 (6800), $\lambda_{min}^{pH\ 14}$ 244 (1600); pmr (DMSO-d_6) δ 2.74 (4, m, CH_2CH_2), 3.47 (2, very broad singlet, H_2O), 5.38 (1, s, H-5), 10.75 (2, broad singlet, NH's). The presence of 1 mol of H_2O supports the analytical data.

Anal. Calcd for $C_6H_7N_2O_5SNa \cdot H_2O$: C, 27.70; H, 3.48; N, 10.76; S, 12.32. Found: C, 27.43; H, 3.35; N, 10.62; S, 12.23.

Polymerization of 6-Vinyluracil.—To a suspension of 0.400 g (0.0029 mol) of 6-vinyluracil in 2 ml of dried acetonitrile was added 1 ml of *N,O*-bis(trimethylsilyl)acetamide (BSA). The container was sealed and the mixture was stirred while being heated at 50° until it became clear. Most of the solvent was then evaporated under a stream of dry nitrogen and excess BSA was removed under vacuum. The syrupy residue was dissolved in 5 ml of dry dioxane, and 10 mg of azobisisobutyronitrile was added. The clear solution was placed in a sealed tube and heated

(15) T. B. Johnson and L. H. Chernoff, *J. Amer. Chem. Soc.*, **36**, 1742 (1914); K. L. Nagpal, P. C. Jain, P. C. Srivastava, M. M. Dhar, and N. Anand, *Indian J. Chem.*, **6**, 762 (1968).

(16) Compound **6** was used as the crystalline monohydrate form. When, on one instance, it was dried and used in the anhydrous form no reaction took place. The apparent need for some water in the reaction mixture is not clear.

(17) P. Rambacher and N. Kaniss, *Angew Chem., Int. Ed. Engl.*, **7**, 383 (1968).

(18) Despite repeated recrystallization of compound **8** the C analysis was consistently lower than the value calculated. Since all other elemental analyses are in excellent agreement with the theoretical values, we feel justified to report the full analysis as presented here.

(19) A large amount of 6-methyluracil was formed as a side product of the reaction when anhydrous conditions and an inert atmosphere were not used.

to 60° for 16 hr. The viscous mixture was then dissolved in 5 ml of 1 *N* NaOH and to the solution was added 15 ml of water. All insoluble material was removed by centrifugation and the polymerized product was precipitated from the clear alkaline solution by neutralization with 5 ml of 1 *N* HCl. The amorphous solid was washed successively with distilled water, methanol, and finally ether. Each washing was performed by suspending the solid in the solvent used and magnetically stirring it for 5 min. It was then recovered by centrifugation and decantation. The product (**13**) obtained after drying *in vacuo* over phosphorus pentoxide weighed 0.386 mg (89% yield based on the analytically determined amount of hydration): uv $\lambda_{max}^{pH\ 1}$ 263 mμ (ε 5600), $\lambda_{min}^{pH\ 1}$ 233 (2000), $\lambda_{max}^{pH\ 11}$ 270 (4600), $\lambda_{min}^{pH\ 11}$ 243 (2000), $\lambda_{max}^{pH\ 14}$ 274 (4500), $\lambda_{min}^{pH\ 14}$ 243 (1900).

Anal. Calcd for $(C_6H_6N_2O_2 \cdot 0.65H_2O)_n$: C, 48.09; H, 4.92; N, 18.70. Found: C, 48.08; H, 4.87; N, 18.69.

1,2-Bis(6-uracilyl)ethene (14).—A solution of anhydrous orotaldehyde **1** (0.560 g, 0.0040 mol) in 20 ml of warm DMF was added slowly to a suspension of **8** (1.69 g, 0.0036 mol) in 60 ml of ethanol). To the clear solution was then added 20 ml of ethanol containing 10 mmol of sodium ethoxide (from 0.240 g of sodium metal). The mixture was heated to reflux for 5 min, cooled to room temperature, and neutralized with glacial acetic acid to pH ~7. The mixture was chilled and the amorphous solid was separated by centrifugation and decantation. It was washed with ethanol and with ether by the same technique. After drying over P_2O_5 the product weighed 0.68 g (76%). Compound **14** was insoluble in all the organic solvents tried and was purified by dissolving in 0.1 *N* NaOH and reprecipitating with an equal volume of 0.1 *N* HCl. The amorphous solid thus obtained was dried over phosphorus pentoxide *in vacuo:* mp >350°; uv $\lambda_{max}^{pH\ 14}$ 340 mμ (ε 14,300), $\lambda_{min}^{pH\ 14}$ 279 (3800), $\lambda_{max}^{pH\ 11}$ 342 (15,900), $\lambda_{mim}^{pH\ 11}$ 283 (3800). (The insolubility of compound **14** precluded accurate uv measurements at lower pH values.)

Anal. Calcd for $C_{10}H_8N_4O_4$: C, 48.39; H, 3.25; N, 22.57. Found: C, 47.86; H, 3.66; N, 22.61.

1,2-Bis(6-uracilyl)ethane (15).—The previous experiment for the preparation of **14** was repeated on a 1-mmol scale. The solid which separated after neutralization (**14**) was resuspended without drying in 50 ml of water and redissolved with the addition of 2.5 ml of 1 *N* NaOH. The solution was hydrogenated at 1 atm over 10% Pd/C. After filtration through Celite, the solution was acidified with glacial acetic acid and the precipitated product was washed with ethanol, then with ether and finally collected by filtration (0.172 g, 76%). Compound **15** (mp >350°) was insoluble in all common organic solvents tried, including hot DMSO and DMF. One purification by the method described for **14** afforded an analytically pure sample of **15**: uv $\lambda_{max}^{pH\ 11}$ 282 mμ (ε 13,500), $\lambda_{min}^{pH\ 11}$ 242 (4200), $\lambda_{max}^{pH\ 14}$ 277 (13,200), $\lambda_{min}^{pH\ 14}$ 244 (4800).

Anal. Calcd for $C_{10}H_{10}N_4O_4$: C, 48.00; H, 4.03; N, 22.39. Found: C, 47.85; H, 4.12; N, 22.36.

6-Methylcytidine by the Mercuric Cyanide–Nitromethane Procedure (18).—A mixture of N^4-acetyl-6-methylcytosine **16**[2b] (1.67 g, 0.010 mol) and mercuric cyanide (5.1 g, 0.020 mol) was suspended in 1 l. of redistilled nitromethane and the mixture was magnetically stirred and heated to reflux. After 100 ml of the solvent had distilled off, a solution of 2,3,5-tri-*O*-benzoyl-D-ribofuranoyl bromide (from 10.5 g of the 1-*O*-acetyl derivative, 0.020 mol) in 100 ml of dry benzene was added slowly to the refluxing mixture. All solids had dissolved by the end of the addition. Heating of the clear solution was continued for 2 hr. After the reaction mixture was cooled, 0.40 g of crystalline unreacted N^4-acetyl-6-methylcytosine precipitated and was recovered by filtration. The clear filtrate was evaporated to dryness and the residue was partitioned between chloroform and a 30% aqueous potassium iodide solution. The organic layer was washed with water, dried over anhydrous sodium sulfate, and evaporated to dryness. The syrupy residue was then dissolved in 200 ml of hot methanol containing sodium methoxide (from 0.100 g of sodium metal). The solution was heated to reflux for ~15 min, cooled to ambient temperature, and neutralized with glacial acetic acid. Evaporation to dryness afforded a syrup which was freed from methyl benzoate by two extractions with ether. The residue crystallized from methanol to afford, in two crops, 1.40 g of 6-methylcytidine (**18**) (71% yield based on unrecovered N^4-acetyl-6-methylcytosine). One recrystallization from methanol gave analytically pure material, mp 236–238° dec (lit.[2b] mp 236–238°). All its other physical constants were identical with those already reported for 6-methylcytidine.

6-Methyluridine (19).—To a solution of **18** (1.20 g, 0.0047 mol) and sodium bisulfite (4.85 g, 0.0467 mol) in 25 ml of water was added acetic acid to bring the pH to ~5. The solution was heated on a steam bath for 2.5 hr, cooled to ambient temperature, and treated with a concentrated aqueous solution of barium hydroxide (8.55 g, 0.500 mol). The mixture was then filtered from the precipitate of barium sulfite and the clear filtrate was passed through a short column of Dowex 50 W (H^+) (170 ml wet volume). After the column was washed free of all uv-absorbing material, the eluate was evaporated to dryness and the residual water was azeotroped with ethanol. Crystallization of the residue from ethanol afforded crystalline 6-methyluridine (0.637 g, 53%) in two crops. The product was identical in all respects with an authentic sample of 6-methyluridine.[2] A study of the rate of deamination of cytidine under the conditions used here indicated that complete conversion to uridine had occurred after only 20 min.

***trans*-3-(6-Uridinyl)acrylic Acid Ethyl Ester (23).**—A solution of 6-methyluridine (1.976 g, 0.0077 mol) in 12 ml of acetic anhydride was heated to reflux for 2 hr. The reaction mixture was cooled to room temperature and treated with 3 ml of water. The solution was again heated for another 20 min and finally evaporated to dryness. The crystalline residue was recrystallized from toluene–ethyl acetate to afford 2.30 g (78%) of pure **2′,3′,5′-tri-*O*-acetyl-6-methyluridine (20)**: mp 152–153°; pmr ($CDCl_3$) δ 2.11 (9, m, $COCH_3$), 2.27 (3, s, CH_3 at C-6), 4.06–4.77 (3, m, H-4′ and H-5′), 5.48–5.90 (4, m, H-1′, H-2′, H-3′, and H-5), 9.70 (1, broad singlet, NH).

To a solution of **20** (1.152 g, 0.0030 mol) in 9 ml of dioxane–acetic acid (11:1) was added 1 g (0.009 mol) of selenium dioxide.[20] The well-stirred suspension was heated to reflux for 14 hr. Tlc of the supernatant solution (chloroform–methanol, 10:1) indicated the initial formation of an intermediate compound with R_f 0.48 (presumed to be the 6-hydroxymethyl derivative). As the reaction proceeded this initial product slowly disappeared as the final product **21** (R_f 0.57) increased in concentration. This aldehyde was detected by its absorbance under uv light and by spraying with a solution of dinitrophenylhydrazine hydrochloride. The mixture was diluted with benzene and filtered. The filtrate was evaporated to dryness and the residue was partitioned between water and chloroform. The organic layer was then dried over sodium sulfate. A small sample of the crude product was chromatographed on a 20 × 20 cm silica gel PF_{254} (2 mm) plate and the band corresponding to the aldehyde was eluted with ethyl acetate. After evaporation of the eluate and drying of the residue *in vacuo*, the pure sample of **21** was used for pmr measurements: pmr ($CDCl_3$) δ 2.10 (9, s, $COCH_3$), 4.09–4.61 (3, m, H-4′ and H-5′), 5.33–6.02 (2, m, H-2′ and H-3′), 6.34 (1, s, H-5), 6.52 (1, d, H-1′), 9.53 (1, s, CHO), 10.00 (1, broad singlet, NH), $J_{1',2'}$ = 2.5–3.0 Hz. Evaporation of the dried chloroform solution afforded **21** as a syrup which was dissolved immediately in 5 ml of dry DMF and treated with 1.22 g (0.0035 mol) of carboxymethylenetriphenylphosphorane under dry nitrogen. Tlc of the mixture after standing at room temperature for 16 hr indicated completion of the reaction. An excess of the phosphorane was destroyed by addition of 1 ml of 40% aqueous formaldehyde and the mixture was evaporated to dryness. The crude residue **22** was dissolved in 50 ml of ethanol containing sodium ethoxide (from 200 mg of sodium metal) and the deacetylation mixture was left standing overnight at room temperature. The stiff gelatinous mass was first diluted with an equal volume of ethanol and then treated with an excess of Dowex 50 W (H^+) with vigorous stirring. Solids dissolved as neutralization proceeded. After filtration, the clear solution was evaporated to dryness and the residue which contained the title compound **23** together with some triphenylphosphine oxide was freed of the latter by extraction with benzene and then ether. Tlc indicated the presence of **23** as the major product together with a minor component (possibly the cis isomer). Crystallization of the syrupy residue from ethyl acetate gave 0.189 g of **23** as white, stout prismatic crystals in three crops, mp 152–156°. The mother liquor was applied to two 20 × 20 cm silica gel PF_{254} (2 mm) plates which after chromatography in chloroform–methanol (10:1) and elution of the appropriate band with ethanol–

(20) Selenium dioxide (99%) was purchased from J. T. Baker Chemical Co., Phillipsburg, N. J.

ethyl acetate (20:80) yielded 0.220 g of pure **23** as a homogenous syrup (40% total yield). This syrup crystallized from ethanol as small aggregates of white, fibrous needles (110 mg in two crops, mp 152–156°). The two crystalline forms obtained for product **23** had identical physical properties (uv, pmr, analysis, tlc) except for their ir spectra (KBr). It was found possible to convert one form (prisms) to the other (fibers) by redissolving crystals of the former in hot ethanol and seeding the solution with crystals of the latter. The ir spectrum was found to have changed accordingly: uv $\lambda_{max}^{pH\ 1}$ 290 mμ (ε 7300), $\lambda_{min}^{pH\ 1}$ 259 (4600), $\lambda_{max}^{pH\ 11}$ 297 (6400), $\lambda_{min}^{pH\ 11}$ 264 (5200); pmr (DMSO-d_6) δ 1.27 (3, t, CH_3), 3.20–4.55 (7, m, H-2′, H-3′, H-4′, H-5′, and $COCH_2$), 4.60–5.40 (3, two doublets and one triplet, all exchangeable in D_2O, sugar OH's), 5.63 (1, d, H-1′), 5.97 (1, s, H-5), 6.61 and 7.54 (2, two AB doublets, CH=CH), 11.49 (1, broad singlet, NH), $J_{1',2'}$ = 5.0 Hz, J_{trans} = 16.0 Hz.

Anal. Calcd for $C_{14}H_{18}N_2O_8$: C, 49.12; H, 5.30; N, 8.18. Found: C, 49.01; H, 5.30; N, 8.09.

Registry No.—**2**, 36807-59-7; **3**, 13345-12-5; *cis*-**4**, 36807-60-0; *trans*-**4**, 36807-61-1; **5**, 22724-20-5; **5** phenylhydrazone, 36803-37-9; **7**, 18592-13-7; **8**, 36803-39-1; **9**, 36803-40-4; **10**, 15043-03-5; **11**, 36803-42-6; **12**, 36803-43-7; **13**, 36812-98-3; **14**, 36803-44-8; **15**, 36803-45-9; **20**, 36807-62-2; **21**, 36807-63-3; **23**, 36806-64-4.

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Interconversions of Hexofuranosyl Nucleosides. IV. Synthesis of Nucleosides Derived from 6-Deoxy-L-glucose[1]

LEON M. LERNER

Department of Biochemistry, State University of New York, Downstate Medical Center, Brooklyn, New York 11203

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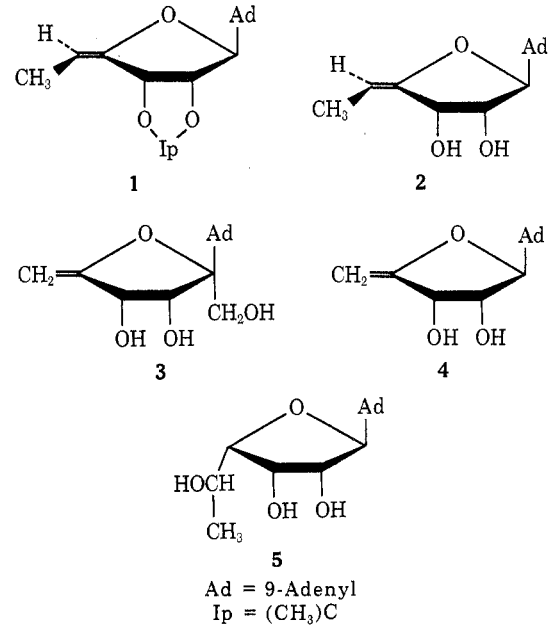
Acetolysis of 6-deoxy-1,5-di-*O*-acetyl-2,3-*O*-isopropylidene-L-mannofuranose (**6**) in 10:1 acetic acid–acetic anhydride followed by reaction of the crude product with 6-benzamidochloromercuripurine and titanium tetrachloride in refluxing 1,2-dichloroethane, gave 9-(6-deoxy-β-L-glucofuranosyl)adenine (**8**) and 9-(6-deoxy-α-L-mannopyranosyl)adenine (**7**) in a ratio of 4:1, after removal of blocking groups. Similarly, acetolysis of 6-deoxy-2,3-*O*-isopropylidene-5-*O*-*p*-toluenesulfonyl-L-mannofuranose under the same conditions, followed by formation of the blocked nucleoside, afforded crystalline 9-(6-deoxy-2,3-di-*O*-acetyl-5-*O*-*p*-toluenesulfonyl-β-L-glucofuranosyl)adenine (**9**) and not 9-(6-deoxy-2,3-di-*O*-acetyl-5-*O*-*p*-toluenesulfonyl-α-L-mannofuranosyl)adenine, as had been previously reported. Acetolysis of **6**, removal of the blocking groups with base, and acetylation of the free sugars gave the crystalline anomeric 6-deoxy-1,2,3,4-tetra-*O*-acetyl-L-glucopyranoses (**11** and **12**) in good yield. 6-Deoxy-L-glucose was prepared in 62% yield from the β anomer **11**. Both **11** and the α anomer **12** were converted to 6-deoxy-2,3,4-tri-*O*-acetyl-α-L-glucopyranosyl bromide (**14**) by reaction with hydrogen bromide in acetic acid. The nucleoside, 9-(6-deoxy-β-L-glucopyranosyl)adenine, was prepared by reaction of **14** with 6-benzamidochloromercuripurine in boiling xylene followed by removal of the blocking groups with sodium methoxide.

In the previous article in this series[2] the synthesis of 9-(5,6-dideoxy-2,3-*O*-isopropylidene-β-D-*erythro*-hex-4-enofuranosyl)adenine (**1**) (Chart I) was described. The deblocked nucleoside **2** was of interest because of its structural relationship to the nucleoside antibiotic, decoyinine (**3**), and to a biologically active analog of **3**, 9-(5-deoxy-β-D-*erythro*-pent-4-enofuranosyl)adenine (**4**).[3] However, removal of the isopropylidene group of **1** under the various acidic conditions attempted resulted in a complete degradation of the nucleoside because of its acid-unstable enol ether structure. The same problem arose in the preparation of **3** and **4**, but this was solved with use of the more acid-labile ethoxymethylidene blocking group in place of the isopropylidene group.[3] Therefore, a decision was made to prepare an alkoxymethylidene derivative of **2** with the expectation that this blocking group could be removed under conditions that would not hydrolyze the *N*-glycosyl bond.[4] To do this it was necessary to prepare alkoxymethylidene derivatives of 9-(6-deoxy-α-L-mannofuranosyl)adenine (**5**); therefore, a large quantity of **5** was required. Two routes leading to **5** have been reported,[5,6] but neither one is straightforward,

CHART I



Ad = 9-Adenyl
Ip = $(CH_3)C$

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(2) L. M. Lerner, *J. Org. Chem.*, **37**, 477 (1972).

(3) J. R. McCarthy, R. K. Robins, and M. J. Robins, *J. Amer. Chem. Soc.*, **90**, 4993 (1968).

(4) C. A. Dekker and L. Goodman in "The Carbohydrates," Vol. IIA, W. Pigman and D. Horton, Ed., Academic Press, New York, N. Y., 1970, p 1.

(5) B. R. Baker and K. Hewson, *J. Org. Chem.*, **22**, 966 (1957).

(6) L. M. Lerner and Y. Y. Cheng, *Carbohyd. Res.*, **14**, 297 (1970).