potassium hydroxide and letting the mixture stand at 20° for 9 days. The dark solution was neutralized with carbon dioxide, treated with decolorizing carbon, extracted with chloroform, deionized by passage through Amberlite IR-120 and Duolite A-4 ion-exchange columns, and the neutral solution concentrated to a sirup that weighed 4.3 g. This sirup was fractionated on a cellulose column with benzene–ethyl alcohol–water mixtures as eluent. Two orcinol-positive fractions were obtained: the first (1.1 g.) yielded 0.55 g. of crystalline 2,7-anhydro-β-D-*manno*-heptulopyranose, the second (1.4 g.) 0.65 g. of D-*gluco*-heptulose.

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Nucleosides. XXIII. 2′,5′- and 3′,5′-Epoxides of Pentofuranosyluracils[1-3]

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Treatment of 1-(5′-*O*-mesyl-2′,3′-epoxy-β-D-lyxosyl)uracil (Ib) with sodium benzylate effects epoxide opening with the formation of 1-(2′,5′-epoxy-3′-*O*-benzyl-β-D-arabinosyl)uracil (X). Hydrogenation of X using palladium–charcoal catalyst gave 1-(2′,5′-epoxy-β-D-arabinofuranosyl)uracil (II). As a proof of structure, epoxide II was synthesized from 1-(5′-*O*-mesyl-β-D-arabinofuranosyl)uracil (VIIb). Both the arabino nucleoside VIIb and 1-(5′-*O*-mesyl-β-D-xylofuranosyl)uracil (VIIIb) were synthesized by opening the epoxide ring of the 5′-*O*-mesyl 2′,3′-epoxide Ib with aqueous sulfuric acid. As proof of structure, arabino nucleoside VIIb was converted to the known tri-*O*-mesylarabinosyluracil. The 5′-mesylated nucleosides VIIb and VIIIb reacted intramolecularly in aqueous alkali, and yielded as sole products 1-(2′,5′-epoxy-β-D-arabinofuranosyl)uracil (II) and 1-(3′,5′-epoxy-β-D-xylofuranosyl)uracil (III), respectively. Hydrolysis of 5′-*O*-mesyl- and 5′-iodo-arabinosyluracils (VIIb and XIX) under slightly acid conditions gave 2,2′-anhydroarabinosyluracil (XXI), together with the 2′,5′-epoxide II and 1-β-D-arabinofuranosyluracil (VIIa). The novel conversion of VIIb and XIX to the 2,2′-anhydro nucleoside XXI is postulated as occurring *via* a 2,5′-anhydroarabinosyluracil intermediate. Hydrolysis of 5′-*O*-mesylxylosyluracil (VIIIb) under the same conditions as VIIb gave 2,3′-anhydroxylosyluracil (XXII), together with the 3′,5′-epoxide III and 1-β-D-xylofuranosyluracil (VIIIa). It is postulated that the anhydro nucleoside XXII arises by attack of the 3′-hydroxyl group at C-2 of a 2,5′-anhydroxylosyluracil intermediate.

Recent reports have described the syntheses of a wide variety of uracil nucleosides from trimesyloxyuridine.[4-8] Among these compounds 1-(2′,3′-epoxy-β-D-lyxofuranosyl)uracil and its 5′-mesyloxy derivative (I, Figure 1) have been shown to be useful intermediates in the preparation of other 1-β-D-aldopentofuranosyluracil analogs of potential biological interest.[6] Of the six possible epoxypentofuranosyluracils (I–VI, Figure 1) only one, the 2′,3′-epoxide I of the *lyxo* configuration, has been reported.[9] The present paper describes the syntheses and properties of 1-(2′,5′-epoxy-β-D-arabinofuranosyl)uracil (II) and 1-(3′,5′-epoxy-β-D-xylofuranosyl)uracil (III) from the 2′,3′-epoxide Ib. It was anticipated that these epoxides would be valuable intermediates in the syntheses of biologically active pyrimidine nucleosides. Epoxides IV and V will be reported in the next paper.[10]

Epoxypentofuranosides in which the epoxide ring is a propylene or butylene oxide type have been reported. These dicyclic furanosides were synthesized by the intramolecular displacement of a 5-sulfonyloxy group by a 2- or 3-alkoxide ion, where the sulfonyloxymethyl group and the attacking alkoxide ion were in *cis* relationship. The known examples of these are methyl-(ethyl) 2,5-epoxy-α-L-arabinofuranoside prepared by Cifonelli and associates,[11] and 1-[2′-deoxy-3′,5′-epoxy-β-D-lyxo(xylo)syl]thymine synthesized by Horwitz and co-workers.[12] Levene and Raymond[13] have described the conversion of 1,2-*O*-isopropylidene-5-*O*-tosyl-D-xylose by sodium methoxide to 1,2-*O*-isopropylidene-3,5-epoxy-D-xylose.

The synthesis of the 2′,5′- and 3′,5′-epoxy nucleosides (II and III) required the availability of 1-(5′-*O*-Ms-β-D-arabinofuranosyl)uracil (VIIb) and 1-(5′-*O*-Ms-β-D-xylofuranosyl)uracil (VIIIb), respectively (Figure 2). The obvious starting material for the arabino nucleoside VIIb was the 5′-*O*-mesyl 2′,3′-epoxy derivative Ib. The opening of the epoxide linkage in Ib with various nucleophilic reagents under basic or acidic conditions was expected to yield products with predominently the *arabino* configuration *via* nucleophilic attack at C-3′.[6,14] A second reaction, the replacement of the 5′-mesyloxy group of Ib by

(1) This investigation was supported in part by funds from the National Cancer Institute, National Institutes of Health, U. S. Public Health Service (Grant CA 03190-08).

(2) A preliminary report has appeared: see I. L. Doerr, J. F. Codington, and J. J. Fox, Abstracts, 145th Meeting of the American Chemical Society, New York, N. Y., Sept. 1963, p. 19D.

(3) In accordance with the suggestion (see ref. 6), the term "epoxy" is used to refer to an ether linkage in the sugar moiety. The term "anhydro" (as "anhydro nucleoside") refers to an oxygen bridge between the C-2 of the pyrimidine and C-2′, C-3′, or C-5′ of the sugar moiety.

(4) J. F. Codington, R. Fecher, and J. J. Fox, *J. Am. Chem. Soc.*, **82**, 2794 (1960).

(5) R. Fecher, J. F. Codington, and J. J. Fox, *ibid.*, **83**, 1889 (1961).

(6) J. F. Codington, R. Fecher, and J. J. Fox, *J. Org. Chem.*, **27**, 163 (1962).

(7) J. F. Codington, I. L. Doerr, and J. J. Fox, *ibid.*, **29**, 558 (1964).

(8) J. F. Codington, I. L. Doerr, and J. J. Fox, *ibid.*, **29**, 564 (1964).

(9) The transient existence of 1-(2′3′-epoxy-β-D-ribosyl)uracil (VI) has been postulated under various alkaline and acid conditions. See ref. 7 for leading references.

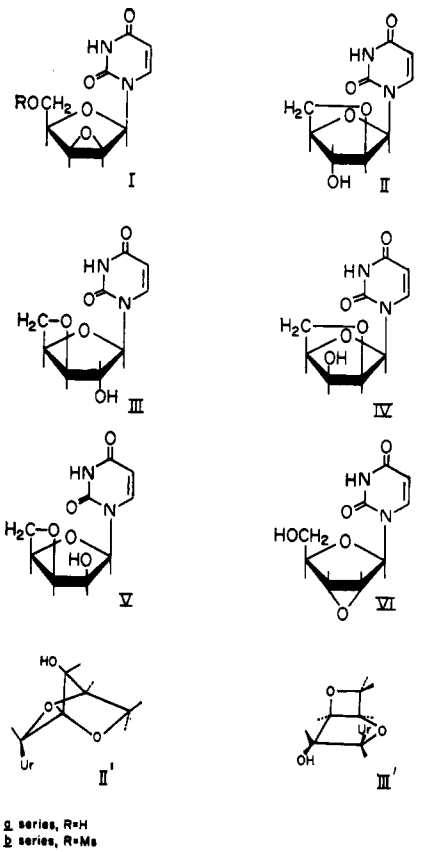
(10) J. F. Codington, I. L. Doerr, and J. J. Fox, *J. Org. Chem.*, **30**, 476 (1965).

(11) M. Cifonelli, J. A. Cifonelli, R. Montgomery, and F. Smith, *J. Am. Chem. Soc.*, **27**, 121 (1955).

(12) J. P. Horwitz, J. Chua, J. A. Urbanski, and M. Noel, *J. Org. Chem.*, **28**, 942 (1962).

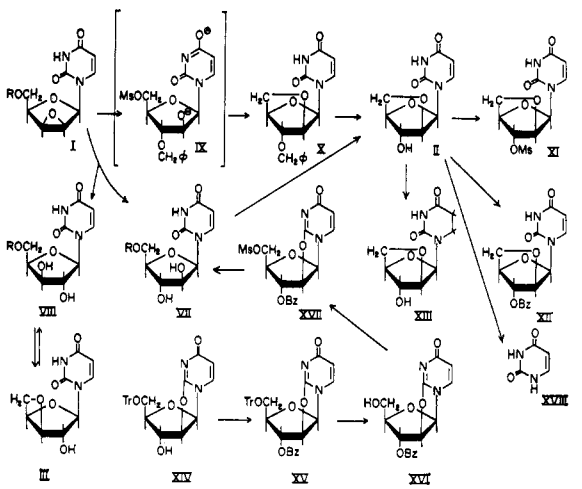
(13) P. A. Levene and A. L. Raymond, *J. Biol. Chem.*, **102**, 331 (1933).

(14) (a) B. R. Baker, R. E. Schaub, and J. H. Williams, *J. Am. Chem. Soc.*, **77**, 7 (1955); (b) B. R. Baker and R. E. Schaub, *ibid.*, **77**, 5900 (1955); (c) W. W. Lee, A. Benitez, L. Goodman, and B. R. Baker, *ibid.*, **82**, 2648 (1960); (d) E. Reist and L. Goodman, *Biochemistry*, **3**, 15 (1964).



a series, R=H
b series, R=Ms

Figure 1.



a, R = H
b, R = Ms
Ms = methanesulfonyl, Bz = benzoyl, Tr = triphenylmethyl

Figure 2.

the nucleophilic agent might also be envisioned as competing with the epoxide-opening reaction.

The reaction of Ib with nucleophiles under basic conditions was investigated. In aqueous 1 *N* sodium hydroxide at room temperature overnight, Ib was recovered. Next, Ib was treated with benzylate ion, a stronger nucleophile than hydroxide ion. When the 5′-*O*-mesyl-2′,3′-epoxide Ib was treated (Figure 2) with an excess of sodium benzylate in benzyl alcohol at 96° for 3 hr., cleavage of the 2′,3′-epoxide ring occurred. From the reaction mixture a pure sulfur-free benzylated epoxy nucleoside X was isolated in 21% yield. The benzyl group of X was removed by catalytic hydrogenation affording 1-(2′,5′-epoxy-β-D-arabinofuranosyl)uracil (II), $[\alpha]^{25}D$ +193°, in good yield. Mesylation or benzoylation of II gave a mono-*O*-mesyl derivative XI and a mono-*O*-benzoyl derivative XII, respectively, in good yield, indicating the presence of a free hydroxyl group in II. Hydrogenation of II using rhodium-on-alumina catalyst afforded 1-(2′,5′-epoxy-β-D-arabinosyl)-5,6-dihydrouracil (XIII) in good yield. The reaction pathway which best explains the formation of the 2′,5′-epoxide X involves a 1-(5′-*O*-mesyl-3′-*O*-benzylarabinosyl)uracil intermediate (dianionic form, IX). Under the reaction conditions described above, intermediate IX reacts intramolecularly by displacement of the 5′-mesyloxy group by the 2′-anionoid to form X. Proof of II as the 2′,5′-epoxide (rather than the 3′,5′-epoxide which could have occurred by benzylate ion attack at C-2′ [15] of Ib) was obtained by the conversion of 5′-*O*-mesylarabinosyluracil (VIIb, Figure 2) quantitatively in aqueous base to the 2′,5′-epoxide, and the corresponding synthesis of the 3′,5′-epoxide III by treatment of 5′-*O*-mesylxylosyluracil (VIIIb, Figure 2) in aqueous base.

The synthesis of VIIb and VIIIb from Ib in aqueous acid required a study of the epoxide ring opening which would be expected to give nucleosides containing the *trans*-α-glycol grouping. In order to anticipate the probable course of aqueous acid cleavage of the 5′-mesyloxy epoxide Ib, a similar cleavage was carried out on the more readily evaluated unsubstituted epoxide Ia. Reflux of 1-(2′,3′-epoxy-β-D-lyxofuranosyl)uracil (Ia)[6] in 2 *N* sulfuric acid for 1 hr. resulted in complete epoxide cleavage and gave the known *trans*-α-glycol nucleosides, 1-(β-D-arabinofuranosyl)uracil (VIIa)[16] and 1-(β-D-xylofuranosyl)uracil (VIIIa),[4] in 85 and 15% yields, respectively, determined by rotational data and paper electrophoretic measurements (borate buffer, pH 6).[17] No glycosyl cleavage was observed during this epoxide opening. It was expected that an acid-catalyzed epoxide opening of the 5′-*O*-mesyl epoxide (Ib) would give 5′-*O*-mesylarabino and -xylo isomers (VIIb and VIIIb) in proportions (85:15) similar to those observed for epoxide Ia.

The 5′-*O*-mesyl 2′,3′-epoxide (Ib) was refluxed for 1 hr. in 2 *N* sulfuric acid. On analysis of the reaction, a recovery of 40–50% starting material indicated that a 5′-mesyloxy group makes the epoxide ring less susceptible to cleavage. When Ib was treated with stronger acid, 3 *N* sulfuric acid, for 1 hr. at reflux, an increase in the rate of epoxide opening was observed. However, the reaction of mesylated Ib in aqueous acid was more complicated than that of unsubstituted epoxide Ia. In addition to the two desired compounds, (Figure 2) VIIb (isolated in 40–50% yield) and VIIIb (isolated in 10% yield), some uracil (XVIII), 1-β-D-(arabinofuranosyl and xylofuranosyl)uracil (VIIa and VIIIa), and starting material (Ib) were also detected. It is probable that uracil, VIIa, and VIIIa arise from further hydrolysis of VIIb and VIIIb under these reaction conditions, since it was subsequently found

(15) The C-2 opening of a 2,3-epoxypentofuranoside with thiobenzylate ion has been reported: G. Cassini and L. Goodman, *J. Am. Chem. Soc.*, **86**, 1427 (1964).

(16) D. M. Brown, A. R. Todd, and S. Varadarajan, *J. Chem. Soc.*, 2388 (1956).

(17) M. P. Gordon, O. M. Intrieri, and G. B. Brown, *J. Am. Chem. Soc.*, **80**, 5161 (1958).

TABLE I

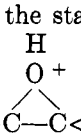
SUMMARY OF THE PRODUCTS FROM THE HYDROLYSES OF 1-(5′-SUBSTITUTED-β-D-PENTOFURANOSYL)URACILS

Reactant	Reflux time, hr.	Hydrolytic agent	Products and their relative amounts[a]
VIIb	14	HOH (~ph 5)	2′,5′-Epoxide (II, 55%)[b] Arabinosyluracil (VIIa, 22%)[c] 2,2′-Anhydroarabino (XXI, 16%)[c] Starting material (VIIb, 7%)
XIX	28.5	HOH (~pH 5)	2′,5′-Epoxide (II, 65%)[b] Arabinosyluracil (VIIa, 6%)[c] 2,2′-Anhydroarabino (XXI, 4%)[c] Starting material (XIX, 25%)
XIX	0.25	Ag^+–HOH	2′,5′-Epoxide (II, 44%)[b] Arabinosyluracil (VIIa, 37%)[b] 2,2′-Anhydroarabino (XXI, 19%)
VIIIb	17.5	HOH (~pH 5)	Xylosyluracil (VIIIa, 58%)[c] 3′,5′-Epoxide (III, 6%)[c] 2,3′-Anhydroxylo (XXII, 5%)[c] Starting material (VIIIb, 31%)
VIIb	1.5	3 *N* sulfuric acid	Uracil (45%) Arabinosyluracil (VIIa, 18%)[c] Starting material (37%)
VIIIb	1.5	3 *N* sulfuric acid	Uracil (65%) Xylosyluracil (VIIIa, 6%)[c] Starting material (29%)

[a] The relative amounts of products were determined by paper chromatographic separation, elution, and ultraviolet spectroscopy. [b] Product isolated. [c] Products evaluated further by paper electrophoresis and paper chromatography.

that in hot 3 *N* sulfuric acid VIIb or VIIIb hydrolyze to uracil and, respectively, to VIIa or VIIIa (*vide infra* Table I). The major arabino component VIIb precipitated when the reaction mixture was cooled and was separated from small amounts of starting material (Ib) by Celite partition chromatography.[18] By subjecting the mother liquor to the same chromatographic procedure, crystalline 5′-*O*-mesyl-xylosyluracil (VIIIb) was separated from the other components (*e.g.*, VIIb, uracil, VIIa, and VIIIa). Each of the 5′-mesyloxy nucleosides (VIIb and VIIIb) consumed 1 equiv. of metaperiodate/mole slowly (5 days), indicating the presence of *trans* vicinal hydroxyls.[19] The mesylated arabino nucleoside VIIb was converted *via* methanesulfonyl chloride–pyridine to 1-(2′,3′,5′-tri-*O*-mesyl-β-D-arabinosyl)uracil which has been previously synthesized in our laboratory.[5] This synthesis constitutes a rigid proof of the *arabino* configuration of VIIb.

The greater stability of the ethylene oxide ring in Ib *vs.* Ia reflected by the decreased rate of epoxide opening in aqueous acid is noteworthy. The decreased rate of reaction appears to be primarily due to the greater electron-withdrawing power of the 5′-mesyloxy group, compared with the 5′-hydroxyl, exerted on the epoxide linkage. This polar (inductive and/or direct) effect[20] would be expected to decrease the stability of



the conjugate acid of the epoxide (>C—C<) which is an intermediate involved in this reaction and thereby diminish the rate of epoxide opening. It should be mentioned also that, although the rate of epoxide opening was decreased in epoxide Ib, the orientation of the opening was the same as in Ia with the arabino isomer VIIIb being formed as the major product *via* epoxide opening at C-3′.

As expected, when 5′-*O*-mesylarabinosyluracil (VIIb) was dissolved in aqueous sodium hydroxide, the 5′-mesyloxy group was intramolecularly displaced by the 2′-anionoid oxygen affording a quantitative yield of 1-(2′,5′-epoxy-β-D-arabinofuranosyl)uracil (II, Figure 2). Compound II was identical with the epoxy nucleoside obtained from the reduction of the benzylated nucleoside X. Therefore, the synthesis, VIIb → II, establishes unequivocally the structure of the epoxy nucleoside which was obtained from X as 1-(2′,5′-epoxy-β-D-arabinofuranosyl)uracil (II).

An alternate synthesis of II *via* VIIb (not isolated) was achieved from the tritylated 2,2′-anhydronucleoside (XIV, Figure 2). Benzoylation of XIV yielded the 3′-*O*-benzoyl derivative XV, which was detritylated with ether–hydrogen chloride to 2,2′-anhydro-1-(3′-*O*-benzoyl-β-D-arabinofuranosyl)uracil (XVI). Mesylation of XVI in pyridine gave the 5′-*O*-mesyl-3′-*O*-benzoyl-2,2′-anhydro nucleoside XVII. Treatment of XVII with excess sodium hydroxide opened the 2,2′-anhydro linkage[21] and removed the 3′-benzoyl group to form VIIb, which under these reaction conditions was converted to the 2′,5′-epoxide II as the sole product.[22] The synthesis of II from 2,2′-anhydro nucleoside XIV further substantiates the structures of VIIb and II prepared from the 2′,3′-epoxide Ib.

Under the same aqueous alkaline conditions that gave the 2′,5′-epoxide II from VIIb, 1-(5′-*O*-mesyl-β-D-xylofuranosyl)uracil (VIIIb) yielded 1-(3′,5′-epoxy-β-D-xylofuranosyl)uracil (III) in good yield. The mechanism of this hydroxide ion dependent reaction involves the intramolecular displacement of the 5′-mesyloxy group by the 3′-anionoid oxygen. The conversion of epoxide III in aqueous acid to 1-β-D-xylofuranosyluracil in excellent yield (see below) confirms the assignment of the 3′,5′-epoxide structure of III and also the 5′-*O*-mesylxylo structure to VIIIb.

Chemical Properties of the 2′,5′-Epoxide II and 3′,5′-Epoxide III.—1-(2′,5′-epoxy-β-D-arabinofuranosyl)uracil (II) and 1-(3′,5′-epoxy-β-D-xylofuranosyl)-uracil (III) were stable in aqueous alkali at room temperature. When II and III were refluxed in water for 24 hr., both nucleosides were recovered unchanged. In hot aqueous acid epoxides II and III were unstable. Nucleophiles were expected to attack the primary carbon (C-5′) in II and III, and, indeed, the 3′,5′-epoxide III on refluxing in 0.2 *N* sulfuric acid for 2 hr. was hydrolyzed to 1-(β-D-xylofuranosyl)uracil (VIIIa). The rate of hydrolysis was dependent on acidity since, in refluxing water (2 ml.) containing 1 drop of glacial acetic acid, only 22% hydrolysis of III to VIIIa occurred in 18 hr. The 3′,5′-epoxide III which contains fused four- (epoxide) and five-(lactol) membered rings is related to bicyclo[3.2.0]heptane as shown in conformation III′ (Figure 1). In conforma-

(18) H. M. Kissman, C. Pidacks, and B. R. Baker, *J. Am. Chem. Soc.* **77**, 18 (1955).

(19) J. J. Fox, N. Yung, J. Davoll, and G. B. Brown, *ibid.*, **78**, 2117 (1956).

(20) M. S. Melvin, "Steric Effects in Organic Chemistry," John Wiley and Sons, Inc., New York, N. Y., 1956, Chapter 13.

(21) The 2,2′-anhydro linkage in 1-(2,2′-anhydro-β-D-arabinofuranosyl)-uracil cleaves in 0.1 *N* sodium hydroxide (2 hr., room temperature) to give only 1-(β-D-arabinofuranosyl)uracil (VIIa).

(22) Horwitz and co-workers[12] explained the formation of 1-(2′-deoxy-3′,5′-epoxy-β-D-lyxosyl)thymine from 5′-*O*-mesyl-2,3′-anhydrothymidine in a similar way.

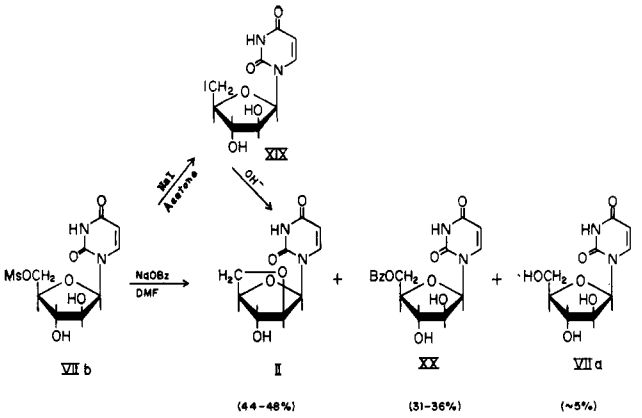


Figure 3.

tion III′ the epoxide ring is in a *cis* relationship to the aglycon, uracil.

The 2′,5′-epoxide II was hydrolyzed completely to uracil (XVIII) and sugar fragment(s) when refluxed in 0.4 *N* sulfuric acid (3 hr.), in contrast to epoxide opening which occurred in III under these conditions. Treatment of II with hydrogen chloride in water-free dioxane (75°)[7] overnight also yielded uracil. Both the anhydrous and aqueous hydrolysates gave a positive aniline–acetate test[23] indicating the presence of furfural, which arose from the sugar moiety of II. The yield of furfural from the aqueous acid hydrolysis of II was only 10%, measured spectrophotometrically, and could not be increased by lengthening the reflux time. 2,5-Epoxy-L-arabinose which is akin to II also yielded furfural on treatment with warm 0.1 *N* sulfuric acid.[10] Attempts to isolate the sugar fragment(s) from the aqueous or anhydrous hydrolysates of II

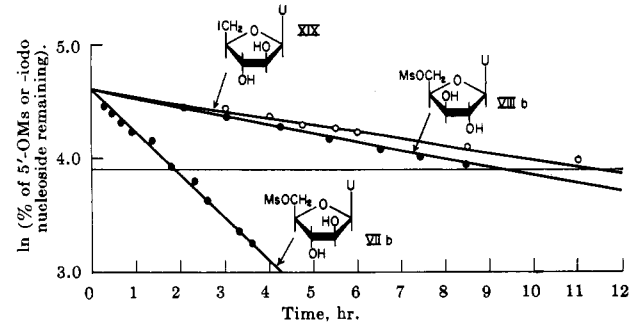


Figure 4.—Graphical expressions for the first-order hydrolysis of 5′-substituted pentofuranosyluracils (~pH 5) at 100°. Intersection of horizontal line represents $t_{1/2}$.

failed. It is extremely interesting that acid conditions (anhydrous and aqueous) cause rupture of the lactol ring rather than the hydrofuranol ring of II. An examination of molecular models (conformation II′, Figure 1) reveals that the 2′,5′-epoxide is a rigid structure of the boat conformation bridged by the C-3′, and is conformationally similar to the 7-norborneol system of Winstein and co-workers.[24] The reaction of II under acid conditions obviously involves protonation of the sugar moiety. However, the mechanism of this reaction cannot be discerned at this time.

Reactions of VIIb under Nonaqueous Conditions.—Displacement of the 5′-mesyloxy group in pyrimidine nucleosides by external nucleophiles is a common reaction. The influence of the pyrimidine moiety in displacement reactions has not been determined, but may be implied from the isolation of 2,5′-anhydro nucleosides. Recently, Letters and Michelson[25] converted 5′-*O*-mesylthymidine to 2,5′-anhydrothymidine by use of sodium *t*-butoxide. The carbonyl group at position 2 of the aglycon participated in this reaction. Studies on 5′-mesyloxy displacement reactions may help to clarify the role of the pyrimidine moiety. The reactions of VIIb with sodium iodide in acetone and with sodium benzoate in N,N-dimethylformamide (DMF) were studied (Figure 3).

The replacement of the 5′-mesyloxy function in VIIb by iodide was expected from previous experiments on 5′-mesylated 1-β-D-pentofuranosyluracils.[4,6] When sodium iodide in acetone was treated with VIIb at 95° for 8 hr., 1-(5′-deoxy-5′-iodo-β-D-arabinofuranosyl)uracil (XIX) was the sole product (68%). Brown and co-workers[26] previously prepared the iodo nucleoside XIX by hydrolysis of 2,2′-anhydro-1-(5′-deoxy-5′-iodo-β-D-arabinofuranosyl)uracil in dilute sulfuric acid. As expected, the reaction of XIX in aqueous sodium hydroxide gave only the 2′,5′-epoxide II (Figure 3).

The replacement of the 5′-mesyloxy function in VIIb by benzoate ion yielding monobenzoate XX was expected from previous work.[4] This reaction was only partially achieved. Treatment of VIIb with one equivalent of sodium benzoate in refluxing DMF for 17 hr. resulted in complete conversion of VIIb to four products. The 2′,5′-epoxide II was the major product (44–48% chromatographically determined, 22% isolated), and 5′-*O*-benzoylarabinosyluracil (XX) was the minor product (31–36% chromatographically determined, 17% isolated). Small amounts of 1-β-D-arabinosyluracil (VIIa, 5% chromatographically determined) and an anhydro nucleoside (presumably 2,2′-anhydroarabinosyluracil,[16] 1%) were also present. These components, VIIa and the anhydro nucleoside, are believed to be artifacts in this reaction, arising probably from the presence of a small amount of water in the DMF. Indeed, VIIa and 2,2′-anhydroarabinosyluracil do arise from the hydrolysis of VIIb (see Hydrolysis Experiments on VIIb and XIX).

Hydrolysis Experiments on VIIb and XIX.—Refluxing VIIb and XIX in water removed, respectively, the 5′-mesyloxy and -iodo groups, as described below in section A. The removal of the 5′-iodo group was also accomplished by treatment of XIX with silver acetate in water as described in B. At the period of time given in column two of Table I, the hydrolyses were halted and the reaction mixtures were analyzed by paper chromatography and electrophoresis. Products were isolated when possible. These experiments gave important information con-

(23) F. Feigl, "Spot Tests in Organic Analysis," Elsevier Publishing Co., New York, N. Y., 1960, page 474.

(24) S. Winstein, M. Shatavsky, C. Norton, and R. B. Woodward, *J. Am. Chem. Soc.*, **77**, 4183 (1955); S. Winstein and E. T. Stafford, *ibid.*, **79**, 505 (1957).

(25) R. Letters and A. M. Michelson, *J. Chem. Soc.*, 1410 (1961).

(26) (a) D. M. Brown, W. Cochran, E. H. Medlin, and S. Varadarajan, *ibid.*, 4873 (1956). (b) The analytical and physical properties of XIX were not reported in ref. 26a.

cerning the removal of 5′ leaving groups in pyrimidine nucleosides of the *arabino* configuration.

A.—VIIb and XIX (0.025 *M*) were refluxed in water, and the liberated acid (MsOH or HI) was neutralized periodically with 0.05 *N* sodium carbonate using methyl red as an indicator. A pH of approximately 5 was kept throughout the hydrolysis. It has been established by Fox and Shugar[27,28] that, in aqueous solutions at pH 5, 1-β-D-aldosyluracils and -thymines exist in the neutral (un-ionized) form (pK_{a_1} for the aglycon = 9–10, pK_{a_2} for sugar hydroxyl(s) = 12–14). In the two reactions, the extent of hydrolysis was determined by titration of the acid liberated. The hydrolyses of VIIb (k_1 = 0.375 hr.$^{-1}$) and XIX (k_1 = 0.062 hr.$^{-1}$) followed good first-order kinetics (Figure 4). The rate of hydrolysis of VIIb was almost six times faster than XIX, indicating that the 5′-mesyloxy group is a far better leaving group than the -iodo group under these conditions.

When 5′-*O*-mesylarabinosyluracil (VIIb) was hydrolyzed in the manner described above, 93% of the methanesulfonic acid was evolved in 14 hr. On analysis, the reaction mixture of VIIb (Table I) was found to contain the 2′,5′-epoxide (55%), 1-β-D-arabinosyluracil (VIIa, 22%), and 2,2′-anhydroarabinosyluracil (XXI, 16%).

The products (II, VIIa, and XXI) from the hydrolysis of 5′-iodo-5′-deoxyarabinosyluracil (XIX) at 73% reaction were the same (Table I) as from the hydrolysis of VIIb. However, the amount of II formed was greater from XIX than from VIIb, as indicated by the ratio II:(VIIa + XXI).[29]

B.—The novel conversion of 5′-*O*-mesyl- or 5′-iodoarabinosyluracils to 2,2′-anhydroarabinosyluracil (XXI) in the above reactions prompted a study of the reaction of the 5′-iodo nucleoside XIX with silver acetate in water. Brown, *et al.*,[26a] reported 1-β-D-arabinosyluracil as the only isolable product when XIX was refluxed in water with 10 equiv. of silver acetate for 15 min. A repetition of this reaction led to the surprising results given in Table I. XIX was converted completely into three products, the 2′,5′-epoxide II (45%), 1-β-D-arabinosyluracil (VIIa, 37%), and 2,2′-anhydroarabinosyluracil (XXI, 19%). The amounts of II, VIIa, and XXI formed from this reaction were about the same as from the hydrolysis of VIIb.

Reaction Mechanism for the Hydrolyses VIIb–HOH, XIX–HOH and XIX–Ag$^+$.—Some preliminary conclusions about the mechanism of 5′-mesyloxy and -iodo group displacement can be drawn from the results of the above hydrolyses. Not only did the three hydrolyses give the same products (II, VIIa, and XXI), but cyclic compounds II and XXI made up over 60% of the total products (Table I). These results suggest that a similar reaction mechanism was operable in all these hydrolyses. From the structure of the cyclic products (II and XXI), the major mesyloxy or iodo displacement mechanism is postulated as one involving

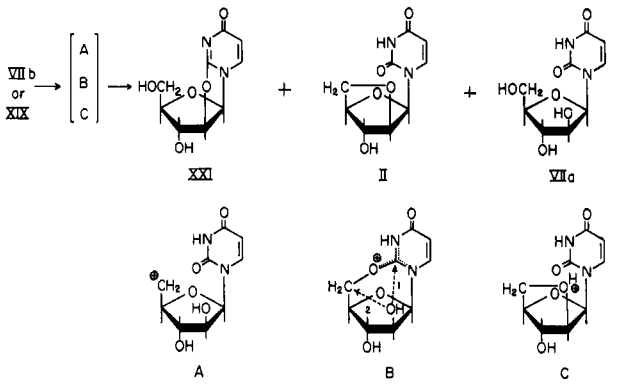


Figure 5.

the participation of the 2′-hydroxyl and 2-carbonyl functions of the nucleosides. More specifically, the hydrolyses of VIIb and XIX are suggested to be complex unimolecular-type[30] displacement reactions in which ions A, B, and C are intermediates (Figure 5). This discussion will not be concerned with the possible bimolecular attack of water on compounds VIIb and XIX which would result in the formation of VIIa.

If the elimination of the C-5′ leaving group in VIIb or XIX is effected by the attack of the 2-carbonyl, then the 2,5′-anhydro nucleoside intermediate B would form. Evidence in favor of cyclic intermediate B is the isolation of 2,2′-anhydroarabinosyluracil (XXI) from the three hydrolyses. The mechanism of 2,2′-anhydro nucleoside formation may be visualized as the intramolecular attack by the 2′-hydroxyl group at the electrophilic pyrimidyl C-2 site of the 2,5′-anhydro resonance hybrid B (arrowed pathway 1, Figure 5). This novel intramolecular conversion of a 2,5′-anhydro nucleoside to a 2,2′-anhydro nucleoside has not been previously reported. Of course, the essential factor for this conversion is the "up" 2′-hydroxyl group in VIIb or XIX.

If the elimination of the C-5′ leaving group in VIIb and XIX is effected by the attack of the 2′-hydroxyl group, then the 2′,5′-epoxide intermediate C would form. Evidence in favor of cyclic intermediate C is the isolation of the 2′,5′-epoxide II from the reactions. An alternate route for II is the intramolecular attack of the 2′-hydroxyl group at the electrophilic C-5′ site of intermediate B (arrowed pathway 2, Figure 5). In keeping with pathway 1 and 2, the 2,5′-anhydro bond of 2,5′-anhydro-2′,3′-isopropylideneuridine is opened by attack of nucleophiles at C-2 and/or C-5′ depending on reagent and reaction conditions.[31,32]

If the elimination of the C-5′ leaving group occurs *without* the participation of neighboring groups, then carbonium A would form. Carbonium A would be expected to form in the first stage of the XIX–Ag$^+$ reaction.[33] Carbonium A can probably be converted to cations B and C by nucleophilic attack of the 2-carbonyl or 2′-hydroxyl functions at C-5′ of A(A ⇆ B and C).

(27) J. J. Fox and D. Shugar, *Biochim. Biophys. Acta*, **9**, 369 (1952).

(28) J. J. Fox, J. F. Codington, N. C. Yung, L. Kaplan, and J. O. Lampen, *J. Am. Chem. Soc.*, **80**, 5155 (1958).

(29) The ratio II:(VIIa + XXI) was 6 for the XIX–HOH reaction, and 1.4 and 0.8 for the VIIb–HOH and XIX–Ag$^+$ reactions, respectively. In this ratio II, VIIa, and XXI refer to the per cents of each component formed if 100% reaction had occurred.

(30) S. Winstein, E. Grunwald' R. E. Buckles, and C. Hanson, *J. Am. Chem. Soc.*, **70**, 816 (1948).

(31) D. M. Brown, D. B. Parihar, A. R. Todd, and S. Varadarjan, *J. Chem. Soc.*, 3028 (1958).

(32) N. K. Kochetkov, E. I. Budowsky, V. N. Shibaev, G. I. Yeliseev, M. A. Grachev, and V. P. Demushklin, *Tetrahedron*, **19**, 1207 (1963).

(33) C. K. Ingold, "Structure and Mechanism in Organic Chemistry," Cornell University Press, Ithaca, N. Y., 1953, page 357.

It should be mentioned that 1-β-D-arabinosyluracil (VIIa) may arise in numerous ways from VIIb and XIX. Intermediates A, B, and C may react at their respective electrophilic sites with water yielding VIIa. Also the product, XXI, may hydrolyze to VIIa.

At this time enough data is not available to specify which intermediate(s) is formed in the rate-determining step in each hydrolysis reaction. Qualitatively, intermediate C appears to be favored in the XIX–HOH reaction, and intermediates A and B in the VIIb–HOH and XIX–Ag^+ reactions. Evidence in favor of this conclusion is the greater abundance of II[29] [expressed by the ratio II:(VIIa + XXI)] in the XIX–HOH reaction than in either of the VIIb–HOH or XIX–Ag^+ reactions.

Hydrolysis of VIIIb.—The hydrolysis of 1-(5′-*O*-mesyl-β-D-xylofuranosyl)uracil (VIIIb) was carried out in a manner similar to the VIIb–HOH and XIX–HOH reactions (*vide supra*, section A). The hydrolysis of VIIIb (k_1 = 0.075) followed first-order kinetics (Figure 4). At 69% reaction, VIIIb was hydrolyzed to three products (Table I): 1-β-D-xylosyluracil (VIIIa, 58%), the 3′,5′-epoxide III (6%), and 2,3′-anhydroxylosyluracil[34] (XXII, 5%). The rate of hydrolysis of 5′-*O*-mesylxylosyluracil (VIIIb) was about five times slower than the arabino analog VIIb indicating that the "up" 3′-hydroxyl group is not so powerful a neighboring group as the "up" 2′-hydroxyl group under these conditions.

The cyclic products III and XXII formed from VIIIb are postulated as arising from a unimolecular-type mesyloxy displacement mechanism involving the 3′-hydroxyl and 2-carbonyl functions. This mechanism is essentially similar to the one proposed for the hydrolysis of VIIb. The elimination of the 5′-mesyloxy group of VIIIb may result in the formation of three intermediates of the *xylo* configuration similar to ions A, B, and C (Figure 5). The 2,3′-anhydro nucleoside XXII is postulated as arising from a 2,5′-anhydroxylosyluracil intermediate (the xylo analog of resonance hybrid B, Figure 5) *via* intramolecular nucleophilic attack by the "up" 3′-hydroxyl group at C-2 of the pyrimidine.

The influence of the pyrimidine moiety in the displacement reactions at C-5′ of pentosyluracils VIIb, VIIIb, and XIX has been demonstrated under neutral hydrolytic conditions. Preliminary data indicate that the replacement of a 5′ leaving group by nucleophiles under nonsolvolytic conditions or solvolytic conditions other than the neutral hydrolyses described above may possibly occur by a similar unimolecular-type mechanism. Therefore, in the complex reaction of 5′-*O*-mesylarabinosyluracil (VIIb) with sodium benzoate–DMF (Figure 3), the products possibly could have arisen *via* ionic intermediates A, B, and C (Figure 5).

Hydrolysis of VIIb and VIIIb in Strong Acid.—As previously discussed, the reaction of 5′-*O*-mesyl-2′,3′-epoxide Ib (Figure 2) with 3 *N* sulfuric acid gave uracil (9–15%)[35] and 1-β-D-arabinosyl- and -xylosyluracils (VIIa and VIIIb, ~3%, see Experimental) in addition to monomesylated nucleosides VIIb and VIIIb. The formation of uracil and nucleosides VIIa and VIIIa from epoxide Ib can be explained as arising from the acid hydrolysis of VIIb and VIIIb.

Compounds VIIb (2.5 mg.) and VIIIb (5 mg.) were refluxed in 3 *N* sulfuric acid (1.5 ml.) for 1.5 hr. The reaction mixtures were neutralized with barium carbonate and analyzed with paper chromatography (system A, upper phase), revealing the presence of three components: starting material (VIIb or VIIIb, R_f 0.4–0.5), uracil (R_f 0.2), and 1-β-D-aldosyluracil (VIIa or VIIIa) (R_f 0.1). The identity of the 1-β-D-aldosyluracil fraction was determined by electrophoresis in borate buffer (pH 6).[4,17] It was found that VIIb and VIIIb had been converted in small amounts to 1-β-D-arabinosyluracil (18%) and 1-β-D-xylosyluracil (6%), respectively (Table I). Large amounts of uracil were formed in both these reactions.

There is an increase in the rate of reaction of VIIb and VIIIb in strong acid over that in neutral solutions. This may be partially explained by a positive ionic strength effect. From analogy to the hydrolysis of VIIIb under neutral conditions, the expected major product from VIIIb in strong acid would be 1-β-D-xylosyluracil (VIIIa, Table I). The other products obtained under neutral conditions, the 3′,5′-epoxide III and the 2,3′-anhydro nucleoside XXII, would be expected to hydrolyze in 3 *N* sulfuric acid predominantly to 1-β-D-xylosyluracil. The large amount of uracil (65%) from VIIIb formed in 3 *N* acid cannot be explained by the unimolecular-type mechanism proposed for the hydrolysis of VIIb and VIIIb under neutral conditions. These results indicate that a reaction mechanism different from that proposed for neutral solutions may be operative in strong acid.

Experimental[36]

Preparation of Chromatographic Systems A and B.[37] **System A.**—To a separatory funnel 1250 ml. of acetone, 500 ml. of chloroform, and 500 ml. of water were added, and shaken. The two-phase system was allowed to stand overnight before use in the partition chromatographic procedure described in the preparation of VIIb and VIIIb. In this chromatographic procedure, the upper and lower phases were the stationary and mobile phases respectively. The lower layer of this two phase system was used as solvent in paper chromatography (ascending technique).

System B.—Acetone–chloroform–water (5:1:1) was used on Schleicher and Schuell paper No. 597 (ascending technique).

1-(3′-*O*-Benzyl-2′,5′-epoxy-β-D-arabinosyl)uracil (X).—To a solution of sodium benzylate obtained from the reaction of 0.40 g. (17 mmoles) of sodium in 15 ml. of benzyl alcohol was added 0.50 g. (1.6 mmoles) of 2′,3′-epoxide Ib. The mixture was heated with stirring for 3 hr. at 86–96° (internal temperature). Benzyl alcohol was removed *in vacuo* below 95°, leaving an amber-colored gum. After thorough trituration with petroleum ether–ether (5:1) and decanting, the residue was freed of organic solvent *in vacuo* and dissolved in 10 ml. of water. Neutralization with acetic acid gave an amber-colored emulsion. Several extractions with ether–petroleum ether (1:1) resulted in a suspension of solid in the aqueous layer. This was collected and triturated in 4 ml. of hot ethanol. After cooling, 0.11 g. (21%) of pale yellow platelets, m.p. 239–242°, were obtained. Crystal-

(34) N. C. Yung and J. J. Fox, *J. Am. Chem. Soc.*, **83**, 3060 (1961).

(35) The amount of uracil formed in this reaction was determined by the ratio of the optical density at 280 and 260 mμ in 1 *N* sodium hydroxide, where 280/260 = 1.49 for uracil and 280/260 = 0.42 for mesyloxy nucleosides.

(36) All melting points were taken on a Thomas–Hoover capillary melting point apparatus and are corrected. Microanalyses were performed by Spang Microanalytical Laboratory, Ann Arbor, Mich. Ultraviolet absorption spectra were run with the Cary Model 15 recording spectrophotometer. Electrophoretic studies were carried out on an E. C. electrophoresis apparatus.

(37) These chromatographic systems were not stable and should be used within 24 hr. after preparation.

lization from ethanol gave colorless micaceous plates, m.p. 241–243°, $[\alpha]^{26}D$ +121° (*c* 0.5, dioxane).

Anal. Calcd. for $C_{16}H_{16}N_2O_5$: C, 60.75; H, 5.10; N, 8.86. Found: C, 60.49; H, 5.05; N, 8.91.

1-(2′,5′-Epoxy-β-D-arabinofuranosyl)uracil (II). Method A.—A solution of 0.14 g. of X in 200 ml. of ethanol was shaken with 0.16 g. of palladium charcoal (5%) in an atmosphere of hydrogen at 20–25° for 115 min. After removal of catalyst, evaporation of solvent gave a crystalline residue. Crystallization from ethanol produced 0.07 g. (70%) of colorless needles, m.p. 249–256° eff. dec., $[\alpha]^{25}D$ +193° (*c* 0.3, water). Ultraviolet absorption spectra at pH 6.9 showed a maximum at 264 mμ (ε 10,700), a minimum at 231 mμ (ε 1900); and in 1 *N* sodium hydroxide showed a maximum at 264 mμ (ε 8460), a minimum at 240 mμ (ε 4270).

Anal. Calcd. for $C_9H_{10}N_2O_5$: C, 47.79; H, 4.42; N, 12.39. Found: C, 48.64; H, 4.75; N, 12.86.

Method B.—A solution of 0.5 g. (1.56 mmoles) of VIIb in 33 ml. of 0.1 *N* sodium hydroxide was allowed to remain at room temperature overnight. The next day, the colorless solution was treated with Dowex 50 (H^+) in order to remove sodium ion. The acid solution was neutralized with 2 *N* triethylamine in ethanol and evaporated *in vacuo* to about 10 ml. The epoxide II precipitated as white needles, 0.35 g. (90%), m.p. 240–243° eff. dec. Recrystallization from ethanol gave colorless needles, m.p. 260–262° eff. dec., $[\alpha]^{23}D$ +190° (*c* 0.30, water). When 2′,3′-epoxide Ib is present in VIIb it contaminates epoxide II (see preparation of VIIb). Epoxide Ib may be removed by washing contaminated II with acetone.

Method C.—When 5′-iodo nucleoside XIX was dissolved in aqueous sodium hydroxide and the reaction mixture was treated similarly as in method B, 2′,5′-epoxide II was obtained in good yield.

Method D.—A solution of 16 mg. of 2,2′-anhydro nucleoside XVII in 3 ml. of 0.15 *N* sodium hydroxide in ethanol–water (1:1) was allowed to stand at room temperature for 2 hr., at which time a constant optical rotation of +200° was obtained. The solution was diluted with 3 ml. of water and applied to a column containing 5 ml. of Dowex 50 (H^+). The ultraviolet-absorbing effluent was evaporated to dryness and triturated with alcohol–ether; 6 mg. (66%) of 2′,5′-epoxide II was obtained, m.p. 250–254° eff. dec. The substance had an identical infrared spectrum as II from method A.

1-(3′,5′-Epoxy-β-D-xylofuranosyl)uracil (III).—To a solution of 0.16 mmole VIIIb in 8 ml. of water was added 2 ml. of 1 *N* sodium hydroxide. The solution was allowed to remain at room temperature overnight. The next day Dowex 50 (H^+) was added to the solution to remove sodium ion. The acid solution was then treated with Dowex 1 (OAc^-) to remove the methylsulfonic acid. The solution was evaporated to dryness and a white residue was obtained. Crystallization from ethanol afforded colorless, flat crystals, 22 mg. (60%), $[\alpha]^{24}D$ −76° (*c* 0.25, water). Ultraviolet absorption properties at pH 6.9 were a maximum at 260.5 mμ (ε 9700), a minimum at 229 mμ (ε 2000); in 1 *N* sodium hydroxide, a maximum at 262.5 mμ (ε 7250), a minimum at 242 mμ (ε 4800).

Anal. Calcd. for $C_9H_{10}N_2O_5$: C, 47.78; H, 4.46; N, 12.38. Found: C, 47.70; H, 4.53; N, 12.71.

1-(2′,5′-Epoxy-3′-*O*-mesyl-β-D-arabinosyl)uracil (XI).—A solution of II (0.11 g., 0.49 mmole) in 8 ml. of pyridine was treated at 0° with 0.09 g. (0.77 mmole) of methanesulfonyl chloride. After remaining at 0–5° for 18 hr., the excess of sulfonyl chloride was destroyed by the addition of 3 drops of ethanol. Solvent was removed *in vacuo*, and the residue was crystallized from 90% ethanol, colorless crystals, 0.14 g. (95% yield), m.p. 196–202° dec. Recrystallization from 90% ethanol gave colorless platelets, 0.10 g., m.p. 196–198° dec., $[\alpha]^{25}D$ +115° (*c* 0.4, acetone).

Anal. Calcd. for $C_{10}H_{12}N_2O_7S$: C, 39.50; H, 3.95; N, 9.20; S, 10.55. Found: C, 39.36; H, 3.96; N, 9.00; S, 10.64.

1-(2′,5′-Epoxy-3′-*O*-benzoyl-β-D-arabinosyl)uracil (XII).—A solution of 0.14 g. (0.62 mmole) of II in 8 ml. of dry pyridine was treated at 0° with 0.120 g. (0.87 mmole) of benzoyl chloride. After remaining at 0–10° overnight, 0.5 ml. of water was added to destroy the excess benzoyl chloride. The reaction mixture was evaporated *in vacuo* to a sirup. Trituration of the sirup with water gave a white solid, 0.14 g. (68%), m.p. 240–245°. Crystallization from ethanol afforded white needles: m.p. 249–250°; $[\alpha]^{24}D$ +15.2° (*c* 0.39, acetone); λ_{max}^{EtOH} 231 and 262 mμ, λ_{min}^{EtOH} 210 and 246 mμ.

Anal. Calcd. for $C_{16}H_{14}N_2O_6$: C, 58.18; H, 4.27; N, 8.48. Found: C, 58.04; H, 4.56; N, 8.34.

1-(2′,5′-Epoxy-β-D-arabinosyl)-5,6-dihydrouracil (XIII).—A solution of 0.15 g. (0.66 mmole) of II in 20 ml. of water was hydrogenated at atmospheric pressure and room temperature using 0.11 g. of rhodium on alumina catalyst. The uptake of hydrogen was quite rapid, the theoretical amount being consumed within 10 min. The catalyst was removed, and the filtrate was evaporated *in vacuo* to dryness. Crystallization from ethanol gave 0.13 g. (87%) of white needles, m.p. 194–195°, $[\alpha]^{23}D$ +72° (*c* 0.37, water). Compound XIII was unstable in aqueous acid and in alkali. The rotation of XIII (+72°) changed immediately to +25° on the addition of 50 λ of 1.5 *N* sulfuric acid. The product(s) formed is unknown. The rotation of XIII decreased in 4.5 hr. to +49° on the addition of 100 λ of 1 *N* sodium hydroxide. A yellow solution was produced which gave a positive *p*-dimethylaminobenzaldehyde–HCl test[38] indicating ureido product(s) had formed.

1-(5′-*O*-Mesyl-β-D-arabinofuranosyl)uracil (VIIb).—A solution of 2.0 g. (6.57 mmoles) of Ib in 18.4 ml. of 3 *N* sulfuric acid was refluxed for 1 hr. On cooling the bright yellow solution, tan-colored needles precipitated (crude VIIb) and were washed with a little cold water, ethanol, and ether: 1.26 g.; m.p. 175–190° darkens, 190–195° dec. (The filtrate from VIIb was saved for the isolation of VIIIb.)

As determined using paper chromatography[39] with the lower phase of system A, crude VIIb contained about 16% of unchanged epoxide Ib. Crude VIIb (0.5 g.) was separated from Ib by passage through a Celite column using solvent system A and the chromatographic procedure described in the preparation of VIIIb (*vide infra*). Pure VIIb was obtained from the fractions containing 256 to 445 ml. of effluent. Evaporation of this effluent to dryness gave a white residue. Crystallization of the residue from water yielded 0.3 g. of colorless needles: m.p. 180–199° darkens, 200–201° dec.; $[\alpha]^{23}D$ +88° (*c* 0.33, water). The ultraviolet absorption spectrum at pH 6.9 had λ_{max} 262 mμ, (ε 10,200), λ_{min} 228 mμ (ε 1630).

Anal. Calcd. for $C_{10}H_{14}N_2O_8S$: C, 37.30; H, 4.37; N, 8.70; S, 9.94. Found: C, 37.21; H, 4.39; N, 8.88; S, 9.75.

1-(2′,3′,5′-Tri-*O*-mesyl-β-D-arabinosyl)uracil[5] was obtained by the reaction of VIIb (0.15 g., 0.466 mmole) in 2 ml. of dry pyridine with methanesulfonyl chloride at 5° overnight. The work-up of this reaction was the same as that previously described.[5] Crystallization of the tri-*O*-mesyl nucleoside from ethanol–water (1:1) gave tan needles, 0.1 g., m.p. 179–182° (previously reported m.p. 179.5–180°), $[\alpha]^{25}D$ +64° (*c* 0.6, dioxane). Rotation of an authentic sample was $[\alpha]^{22}D$ +62° (*c* 0.69, dioxane).[40]

1-(5′-*O*-Mesyl-β-D-xylofuranosyl)uracil (VIIIb).—The filtrate from crude VIIb (see above) contained VIIb, VIIIb, uracil (XVIII), and small amounts of VIIa, VIIIa,[41] and Ib. Removal of the sulfuric acid was accomplished by addition of the filtrate to 11 g. of barium carbonate suspended in 200 ml. of water. When the suspension became neutral to pH paper, the barium salt mixture was filtered off and the filtrate was concentrated to a sirup which was azeotroped with alcohol. The gum obtained was partially dissolved in 60 ml. of acetone at room temperature. The acetone insolubles were mostly uracil and barium salts. The acetone filtrate was concentrated to dryness. A glass was obtained which contained VIIb and VIIIb as the predominant components and was used in the following chromatographic procedure.

The glass was dissolved in 3 ml. of the upper phase (system A) and mixed with 6 g. of Celite. The mixture was immediately packed on to a Celite column (48 × 3 cm.). The column had been previously packed with 90 g. of Celite which was mixed well with 45 ml. of upper phase (system A). The lower phase of system A was passed through the column and 15-ml. effluent aliquots were collected. Fractions 2–4 contained starting ma-

(38) R. M. Fink, R. E. Cline, C. McGaughey, and K. Fink, *Anal. Chem.*, **28**, 4 (1956).

(39) The two spots corresponding to VIIb (R_f 0.6) and Ib (R_f 0.95) were eluted with 2.5 ml. of water. The ultraviolet absorption at 260 mμ of these solutions were determined. This data gave the relative amounts of Ib and VIIb in the mixture.

(40) Previously reported,[5] $[\alpha]^{26}D$ +71° (*c* 0.5, dioxane).

(41) Fractions 28–35 contained uracil. After the uracil was obtained, the Celite column was washed with the stationary phase (upper). Fractions 1–6 contained nucleosides VIIa (9 parts) and VIIIa (1 part). The yield of VIIa and VIIIa from epoxide Ib (0.657 mmole) was 2–3% (spectrophotometrically determined).

terial Ib. Fractions 13–24 (195–360 ml.)[41] contained the 5′-*O*-mesyl nucleosides VIIb and VIIIb which were separated in the following manner.

Owing to the ultraviolet absorption of acetone in the effluent, 2 drops of effluent from each fraction (13–24) were applied to chromatography paper. When dry, the ultraviolet spot was cut from the paper and eluted with 4 ml. of water. The optical density of the solution was determined at 260 mμ and a plot of the O.D. *vs.* the total volume for fractions 13–24 was made. The curve including fractions 13–17 showed a rapid continuous increase in O.D. with a maximum at the 17th fraction. The curve including fractions 17–20 showed a rapid continuous decrease. A break in this curve occurred at fraction 20, and a shoulder appeared. The curve including fractions 20–24 showed a much less rapid O.D. decrease than 17–20. The shoulder at fraction 20 is due to the presence of VIIb. Fractions 13–15 were evaporated; 0.02 g. of solid A was obtained. Fractions 16–19 were evaporated; 0.156 g. of solid B was obtained. Fractions 20–24 were evaporated; 0.058 g. of solid C[42] was obtained. Solids A, B, and C had optical rotations (in water) of +30, +35, and +62°, respectively. Crystallization of solids A and B from 85% ethanol gave 0.11 g. of colorless prisms: m.p. 175° darkens, 185–190° dec.; $[\alpha]^{24}D$ +31° (*c* 1.66, water). The ultraviolet absorption spectrum at pH 6.9 had λ_{max} 261–262 mμ, λ_{min} 230.5 mμ.

Anal. Calcd. for $C_{10}H_{14}N_2O_8S$: C, 37.30; H, 4.37; N, 8.70; S, 9.94. Found: C, 37.43; H, 4.46; N, 8.65; S, 9.96.

Metaperiodate Studies.—The metaperiodate studies on nucleosides VIIb and VIIIb were carried out according to the procedures described in a previous publication.[19] No acid (formic or methylsulfonic acid) was liberated during the study. The consumption of metaperiodate was as follows.

	Moles of IO_4^- consumed/mole				
	2 hr.	54 hr.	3 days	4 days	5 days
VIIb	0.14	0.72	0.87	1.0	1.05
VIIIb	0.14	0.64	0.73	0.94	1.06

1-(5′-Iodo-β-D-arabinofuranosyl)uracil (XIX).—A suspension of 0.30 g. (0.93 mmole) of VIIb in 20 ml. of dry acetone containing 0.28 g. of anhydrous sodium iodide was heated in a sealed glass-lined steel container at 95° for 5 hr. The yellow-colored reaction mixture containing some precipitated sodium mesylate was taken to dryness *in vacuo*. On the addition of water to the orange residue, a pale yellow solid precipitated, 0.22 g., m.p. 183–188° eff. Crystallization of the solid from 20 ml. of water gave colorless rods in 40% yield: m.p. 188–190° eff., 206° evolution of purple fumes; $[\alpha]^{22}D$ +99° (*c* 0.35, water); λ_{max}^{water} 261 mμ, λ_{min} 230 mμ.

Anal. Calcd. for $C_9H_{11}IN_2O_5$: C, 30.53; H, 3.14; I, 35.84; N, 7.91. Found: C, 30.65; H, 3.06; I, 34.55; N, 7.64.

Reaction of VII with Sodium Benzoate–DMF. Isolation of 1-(5′-*O*-Benzoyl-β-D-arabinofuranosyl)uracil (XX) and II.—A solution of 0.109 g. (0.338 mmole) of VIIb in 20 ml. of N,N-dimethylformamide containing 0.054 g. (0.385 mmole) sodium benzoate was refluxed for 17 hr. The pale yellow solution was concentrated *in vacuo* to a sirup. On the addition of water to the sirup, a gum precipitated. The gum was triturated with chloroform, and the monobenzoylated nucleoside XX (18%), m.p. 205–210°, $[\alpha]^{24}D$ +115° (*c* 0.077, 50% ethanol), was collected. The ultraviolet absorption spectrum in water had λ_{max} 262 and 232 mμ (ε 9400 and 12,800, respectively), λ_{min} 249 and 214 mμ (ε 8060 and 10,100, respectively).

Anal. Calcd. for $C_{16}H_{15}N_2O_7$: C, 55.33; H, 4.35; N, 8.07. Found: C, 54.96; H, 4.75; N, 7.80.

Hydrolysis of XX with aqueous potassium hydroxide gave solely 1-β-D-arabinosyluracil (VIIa) as demonstrated by paper electrophoresis (pH 6, borate buffer).[4,17]

The aqueous filtrate from XX was evaporated *in vacuo* to dryness. The residue was triturated with ether, and a yellow solid Y was obtained, 94 mg. (0.229 mmole, 69%).[43] The solid Y was chromatographed (system B), and was found to be a mixture of monobenzoate XX (R_f 0.96), epoxide II (R_f 0.83), spongouridine VIIa (R_f 0.68), and 2,2′-anhydro nucleoside XXI[44] (R_f 0.47). No starting material VIIb (R_f 0.90) was detected. The relative amounts of these compounds were determined by elution (2.5 ml. of water) from the chromatogram followed by an ultraviolet absorption determination. These values were converted into approximate yields of each nucleoside contained in solid Y. The value for XX was adjusted for the amount of XX isolated (*vide supra*). The approximate yields of the nucleoside constituents in the total reaction were XX (31–36%), II (44–48%), and VIIa (~1%). When a small sample of solid Y was hydrolyzed in aqueous sodium hydroxide, the only compounds detected by paper electrophoresis in borate buffers pH 6 and 9 were spongouridine (VIIa) and unchanged epoxide II. The remainder of solid Y was recrystallized from ethanol; 2′,5′-epoxide (II) precipitated in 25% yield, m.p. 260–265° dec. The infrared spectrum was identical with that of authentic II.

Conversion of the 2′,5′-Epoxide II to Uracil (XVIII) in Dilute Acid.—The epoxide II (0.11 g.) was refluxed in 15 ml. of 0.4 *N* sulfuric acid for 3 hr. The ultraviolet properties of the reaction mixture were, in water, maximum at 260 mμ and minimum at 227 mμ; in 1*N* sodium hydroxide, maximum at 276 mμ and minimum at 242 mμ, ratio at 280/260 mμ = 1.5 (for uracil 280/260 mμ = 1.49). The reaction mixture gave a positive test for furfural (aniline–acetate test).[23] The amount of furfural[45] in the reaction mixture was determined by measuring the absorption at 290 mμ in water. The results indicated that 10% of furfural had been formed from II. Therefore after glycosylic cleavage, the sugar moiety of II was *not* completely converted to furfural. The rotation of the reaction mixture was $[\alpha]^{24}D$ −8.2° (*c* 0.7, dilute sulfuric acid), if 2,5-epoxy-D-arabinose[46] is assumed to be formed. The reaction mixture was evaporated *in vacuo* to about 7 ml.; uracil (66%) precipitated, m.p. 330° dec. (no melting point depression with authentic uracil).

An attempt to form a derivative of the sugar residue from II failed. The reaction mixture was neutralized with barium carbonate, filtered, and evaporated *in vacuo* to dryness. The sirupy residue gave a positive Fehling and Benedict test indicating the presence of a reducing sugar. Benzoylation or acetylation of the residue failed to afford a crystalline product. Attempts to form phenylhydrazones from the residue also failed.

Treatment of 1-(2′,3′-Epoxy-β-D-lyxofuranosyl)uracil (Ia)[6] with 2 *N* Sulfuric Acid.—Ia (0.171 g.) was refluxed in 5 ml. of 2 *N* sulfuric acid for 1 hr. The reaction mixture was cooled. The rotation, $[\alpha]^{24}D$ +111° (*c* 0.37), corresponded to a mixture of 85% 1-β-D-arabinosyluracil (VIIa) and 15% 1-β-D-xylosyluracil (VIIIa). These proportions of VIIa and VIIIa were also found on using the following ionophoretic procedure. The reaction mixture was neutralized with barium carbonate and filtered. A 97% recovery of 1-β-d-pentosyluracils was indicated by spectrophotometric methods. No uracil or starting material Ia was present, as determined by paper chromatography (lower phase, system A). The nucleoside solution was concentrated and then made up to a volume of 50 ml. Aliquots of 50 λ were subjected to ionophoretic migration (800 v., 20 ma., 2.5 hr.) in borate buffer, pH 6.[4,17] Two components were present corresponding to VIIa and VIIIa (86:14). The relative proportions were determined spectrophotometrically. 1-β-D-arabinosyluracil (VIIa) was isolated from this reaction in a 62% yield, m.p. 220°.

Conversion of the 3′,5′-Epoxide III to 1-β-D-xylosyluracil[4] (VIIa).—III (6.8 mg.) was refluxed in 2.5 ml. of 0.2 *N* sulfuric acid for 1.5 hr. The reaction mixture was cooled, $[\alpha]^{23}D$ +24° (*c* 0.2) (previously reported[4] for 1-β-D-xylosyluracil, $[\alpha]^{23}D$ +29°). The presence of about 10% uracil was detected in the reaction mixture by the ultraviolet absorption ratio at 280/260 mμ in 1 *N* sodium hydroxide. The reaction mixture was neutralized with barium carbonate and filtered. The filtrate was analyzed by paper chromatography (lower phase, system A), and then by ionophoretic migration (900 v., 24 ma., 2.5 hr.) in borate buffer, pH 6.[4,17] Paper chromatography gave two spots (R_f 0.19 and 0.07) which, on elution, corresponded spectrophotometrically to

(42) Solid C contained a 1:1 mixture of VIIb and VIIIb.

(43) The ultraviolet absorption of the solid Y was determined on a weighed sample. The reasonable assumption was made that a pure mixture of the nucleosides XX, II, and VIIa would have an approximate ϵ_{max} 10,000 at 262 mμ. Based on these calculations the solid Y contained 0.0229 mmole, and accounted for 69% of reactant VIIa.

(44) The identity of this compound is solely based on its identical chromatographic migration (system B) with authentic 2,2′-anhydro nucleoside XXI.

(45) Ultraviolet absorption of furfural in water showed a maximum at 276 mμ (ϵ_{max} 15,100) and a minimum at 243 mμ (ϵ_{min} 1860). The extinction (ε) at 290 mμ was 8660.

(46) The enantiomorph of this sugar, 2,5-epoxy-L-arabinose, has been reported,[11] $[\alpha]^{24}D$ +12°.

uracil (10%) and a pentosyluracil (90%). Ionophoretic migration of the nucleoside spot established the component as 1-β-D-xylosyluracil.

2,2′-Anhydro-1-(5′-*O*-trityl-3′-*O*-benzoyl-β-D-arabinosyl)uracil (XV).—One gram (2.13 mmoles) of XIV was dissolved in 15 ml. of dry pyridine. The solution was cooled to 0° and 2.58 mmoles of benzoyl chloride was added. The reaction mixture was refrigerated overnight, 0.5 ml. of water was added, and the reaction mixture was concentrated *in vacuo* to a thick sirup. On addition of water, solidification occurred. The white solid was dissolved in chloroform, and the chloroform solution was dried and concentrated *in vacuo* to dryness. The residue was crystallized from 20 ml. of ethanol: 0.9 g. (74%), m.p. 225–230°, $[\alpha]^{23}D$ −27° (*c* 0.23, methanol). The ultraviolet absorption spectrum in ethanol showed shoulders at 227 and 250 mμ.

Anal. Calcd. for $C_{35}H_{28}N_2O_6$: C, 73.41; H, 4.93; N, 4.89. Found: C, 73.55; H, 5.32; N, 5.26.

2,2′-Anhydro-1-(3′-*O*-benzoyl-β-D-arabinosyl)uracil (XVI).—XV was suspended in 15 ml. of dry ether. Hydrogen chloride gas was passed through the suspension for 7 min. The reaction flask was sealed and refrigerated for 1 hr. The product was filtered and washed thoroughly with fresh ether. A theoretical yield of a hydrogen chloride salt of XVI was obtained. The salt was decomposed by suspension in water. The product was washed with water until the filtrate gave no test for chloride ion, yield 90%. An analytical sample was prepared by crystallization from ethanol, m.p. 233–235°. The ultraviolet absorption spectrum in ethanol had a maximum at 227 and a shoulder at 255 mμ.

Anal. Calcd. for $C_{16}H_{14}N_2O_6$: C, 58.18; H, 4.27; N, 8.48. Found: C, 58.03; H, 4.24; N, 8.42.

2,2′-Anhydro-1-(5′-*O*-mesyl-3′-*O*-benzoyl-β-D-arabinosyl)uracil (XVII).—Under anhydrous conditions 0.3 g. (0.91 mmole) of XVI was dissolved in 3 ml. of dry pyridine and cooled to 0°. Methanesulfonyl chloride (1 mmole) was added. A blue color formed immediately. The reaction mixture was refrigerated overnight, 0.4 ml. of water was added, and the purple-colored reaction mixture was concentrated to dryness. On the addition of water to the residue, a purple solid precipitated. After filtration the product was washed thoroughly with ethanol, pink solid, yield 50%, m.p. 200–202°. The analytical sample was obtained by crystallization first from acetone, and then from ethanol: micaceous plates, m.p. 198–200°, $[\alpha]^{23}D$ −64° (*c* 0.16, acetone). The ultraviolet absorption spectrum in ethanol had a maximum at 231 and a shoulder at 262 mμ.

Anal. Calcd. for $C_{17}H_{16}O_8N_2S$: C, 50.00; H, 3.95; N, 6.86; S, 7.85. Found: C, 50.04; H, 3.82; N, 6.74; S, 7.76.

Hydrolysis of 1-(5′-*O*-mesyl-β-D-arabinosyl)uracil (VIIb) and 1-(5′-Iodo-5′-deoxy-β-D-arabinosyluracil (XIX). Isolation of 2,2′-Anhydro-1-β-D-arabinosyluracil (XXI), II, and VIIa. A.—VIIb (0.34 mmole) was refluxed in 13 ml. of water with methyl red as an internal indicator. The reaction was titrated periodically with 0.05 *N* sodium carbonate in order to measure the liberated methanesulfonic acid (Figure 4) and also to keep the pH about 5. After a 14-hr. reflux, 94% of the theoretical 1 equiv. of methanesulfonic acid had been evolved. The reaction was cooled, and the solution was chromatographed (system B). The reaction mixture contained four components with identical migration behavior to starting material VIIb (R_f 0.89), the epoxide II (R_f 0.79), 1-β-D-arabinosyluracil (VIIa, R_f 0.65), and the 2,2′-anhydro nucleoside (XXI) (R_f 0.43). Each chromatographic spot was eluted with an equal aliquot of water (2.5 ml.) and the spectral properties of the resulting solutions were determined. The spectra of the four components were identical with those of VIIb, II, VIIa, and XXI. For each component, the optical density values at 260 mμ were converted into relative per cents of VIIb, II, VIIa, and XXI (see Table I).

The reaction mixture was concentrated *in vacuo* to about 3 ml. The 2′,5′-epoxide II precipitated, 17 mg. (23%), m.p. 250–256° eff. The filtrate was treated batchwise with Dowex 50 (H^+), and then Dowex 1 (acetate), to remove the sodium mesylate, and evaporated to about 3 ml. This solution (0.5 ml.) was chromatographed (system B) on Whatman 3MM paper. The XXI-containing strip was eluted with water and evaporated to dryness. On the addition of alcohol, anhydro nucleoside crystals precipitated. This sample had an infrared and ultraviolet spectra essentially superimposable with an authentic sample of XXI.[16] Similarly, when the VIIa-containing strip was eluted, evaporated, and treated with alcohol, crystalline VIIa was obtained. This compound had identical properties with an authentic sample.[16]

B.—XIX (0.15 g.) was refluxed in water (18 ml.) and the hydrolysis mixture was neutralized and analyzed in the manner described above in section A. After 28.5 hr., 75% of the theoretical 1 equiv. of hydroiodic acid was liberated. The reaction mixture was chromatographed (system B) and four components were detected: starting material XIX (R_f 0.95), the epoxide II (R_f 0.74), spongouridine (VIIa, R_f 0.60), and the 2,2′-anhydro nucleoside XXI (R_f 0.38). The relative per cents of each component in the reaction mixture are found in Table I. VIIa- and XXI-containing strips had identical electrophoretic migrations in borate buffers (pH 6 and 9)[4,17] as authentic samples of these compounds. The 2′,5′-epoxide II was isolated in 33% yield on concentration of the reaction mixture *in vacuo* to a small volume.

C.—This experiment is a modification of the one reported by Brown and associates.[26a] XIX (0.2 g.) was heated at 100° with silver acetate (0.8 g.) in water (27 ml.) for 15 min. The reaction mixture was filtered through Celite. Silver ions were removed by hydrogen sulfide and the resulting suspension was filtered through Celite. The filtrate was evaporated *in vacuo* to dryness, and the residue was dissolved in hot ethanol and treated with charcoal. The resulting ethanolic solution was analyzed and found to contain II, VIIa, and XXI in the relative per cents given in Table I. The ethanolic solution was concentrated *in vacuo* to 4 ml., and a crystalline mixture of II and XXI precipitated (28%). The filtrate was concentrated to 2 ml. and compound VIIa precipitated (22%), m.p. 215–219° (authentic sample, m.p. 218–220°). The crystalline mixture of II and XXI was resolved by paper chromatography (Whatman 3MM, system B). The II- and XXI-containing strips gave crystalline compounds on elution. The infrared spectra of compounds II and XXI were superimposable with authentic samples.

Hydrolysis of 1-(5′-*O*-Mesyl-β-D-xylofuranosyl)uracil (VIIIb). Detection of 2,3′-Anhydro-1-β-D-xylosyluracil (XXII), III, and VIIIa.—VIIIb (0.073 mmole) was refluxed in water (2.9 ml.) by the same experimental procedure described for the hydrolysis of VIIb, section A. After a 17.5-hr. reflux, the reaction mixture was chromatographed (system B) and found to contain four components with identical migration behavior to the starting material VIIIb (R_f 0.89), 1-β-D-xylosyluracil (R_f 0.63), the epoxide III (R_f 0.80), and 2,3′-anhydro-β-D-xylosyluracil (R_f 0.39) in the relative per cents given in Table I. The entire hydrolysate was chromatographed on Whatman paper (3MM, system B). The VIIIa-, III-, and XXII-containing strips had identical electrophoretic migrations in borate buffers (pH 6 and 9)[4,17,47] with authentic samples of these compounds. The 3′,5′-epoxide III-containing strip was subjected to hydrolysis in 0.2 *N* sulfuric acid. The neutralized hydrolysate contained one compound which had an identical electrophoretic migration in borate buffer (pH 6) with an authentic sample of 1-β-D-xylosyluracil (VIIIa).

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(47) Personal communication from N. C. Miller. The electrophoretic migrations of 2,2′-anhydro-β-D-arabinosyluracil and 2,3′-anhydro-β-D-xylosyluracil after 2 hr. are +1.9 and −1.0 cm., respectively, in borate buffer (pH 9.2, 80–100 ma., 750 v.) and essentially the same (−1.3 cm.) in borate buffer (pH 6, 20 ma., 900 v.). The ultraviolet spectra of the two anhydro nucleosides are appreciably different.[34,10]