ture was left at 0° for 2 hr. and then allowed to warm to room temperature overnight. The reaction mixture was then cooled in an ice bath, and 10 ml. of water was added dropwise over a period of 1 hr. The reaction mixture was then poured into 500 ml. of ice and water to produce a crystalline solid which upon recrystallization from chloroform–ethanol weighed 4.76 g. (65% over-all yield), m.p. 131–132°, $[\alpha]^{23}D$ +10° (*c* 1.19, chloroform).

Anal. Calcd. for $C_{25}H_{24}O_7S$: C, 64.09; H, 5.16; S, 6.84. Found: C, 63.95; H, 5.18; S, 6.69.

Other Materials.—The benzenesulfonate esters, I,[4] II,[5] III,[6] and IV,[7] were prepared by methods used in the synthesis of the corresponding *p*-toluenesulfonate esters. Data obtained are given in Table III. Compounds VI and VII previously were characterized.[9] Acetonylacetone was fractionally distilled through a 40-cm. Vigreux column. The fraction boiling at 101–102° at 46 mm. or 82° at 23 mm. was collected. Standard solutions (0.100 *M* or 0.080 *M*) of sodium iodide, Baker and Adamson analyzed, dried at 110° overnight, in acetonylacetone were prepared. Standard solutions (0.100 *M* or 0.080 *M* in sulfonate) of each of the benzenesulfonate esters in acetonylacetone were prepared. A standard solution, approximately 0.03 *M*, of silver nitrate, Baker and Adamson analyzed, dried overnight at 110°, was used in Volhard[19] or argentimetric[20] titrations for iodide ion.

Kinetic Studies.—A constant temperature bath containing mineral oil with temperature control of ±0.05° in the vicinity of 75° and of ±0.1° in the vicinity of 125° was used. For a run at a given temperature, nineteen 18 × 150 mm. Pyrex test tubes were used. Aliquots of 5.00 ml. of a standard sulfonate ester solution were introduced at room temperature. Then 5.00 ml. of the standard iodide solution was introduced into each tube at room temperature and immediately immersed into the bath. Tubes were withdrawn at varying intervals and quickly immersed in a mixture of ice and water. The contents of each tube were transferred, using distilled water or acetone to effect quantitative removal, into an erlenmeyer flask and titrated for unreacted iodide.

Acknowledgment.—A University of Utah Research Committee Fellowship to W. J. Teerlink during the summer of 1962 is gratefully acknowledged.

(19) J. R. Caldwell and H. V. Moyer, *Ind. Eng. Chem., Anal. Ed.*, **7**, 38 (1935).

(20) I. M. Kolthoff and E. B. Sandell, "Textbook of Quantitative Inorganic Analysis," 3rd Ed., The Macmillan Co., New York, N. Y., 1952, p. 544.

The Synthesis of Some 5′-Thiopentofuranosylpyrimidines[1]

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The synthesis of 5′-thiothymidine (XIIIa), 2′-deoxy-5-fluoro-5′-thiouridine (XIIIb), 5′-thiouridine (III), and a number of acylated derivatives of XIIIa and XIIIb is described. The tendency of XIIIa, XIIIb, and III to form cyclonucleosides by addition of the thiol group across the uracil carbon–carbon double bond was studied. No evidence could be obtained for cyclonucleoside formation from 5′-thiothymidine (XIIIa) at pH 1 or 7. At pH 13, disulfide formation occurred. The fluoronucleoside (XIIIb) formed disulfide quite rapidly at pH 13. At pH 7, an equilibrium which exists between the open-chain nucleoside (XIIIb) and the cyclonucleoside (XVb) is ultimately displaced to the open-chain form by disulfide formation. At pH 1, the open-chain nucleoside (XIIIb) appears stable. A solution of 5′-thiouridine (III) at pH 7 exists in equilibrium with the cyclonucleoside (IVb). At pH 1, there was no evidence for cyclonucleoside formation, while at pH 13 disulfide formation occurred.

Bannister and Kagan[2a] recently reported on their efforts to prepare 5′-thiouridine (III) and its acetonide (II). They observed[2a] that treatment of 5′-S-acetyl-2′,3′-O-isopropylidine-5′-thiouridine (I) with methanolic ammonia gave, instead of the expected acetonide (II) of 5′-mercaptouridine, a product in which the thiol group of II had added across the uracil double bond to give the isomeric cyclic sulfide (IVa). (See Scheme I.)

The formation of the cyclic sulfide was suggested by the disappearance of the characteristic uridine ultraviolet absorption peak in the 260-mμ region. Their work indicated that, under alkaline conditions, this cyclization was reversible and that the "normal" structure (II) was regenerated. More recently, Chambers and Kurkov[2b] observed ready formation of the cyclonucleoside (IVa) at neutral pH. In one experiment, crystalline II was isolated by rapid acidification of an alkaline solution of II. In subsequent attempts to repeat this, however, invariably the cyclonucleoside (IVa) separated although the presence of acetone 5′-thiouridine in solution was suggested from the ultraviolet spectrum. We recently prepared a series of acylated derivatives of 5′-thiothymidine (XIIIa) and 2′-deoxy-5-fluoro-5′-thiouridine (XIIIb) for another purpose and thought it would be interesting to investigate the possibility of a similar cyclonucleoside formation with the free thiols.

The synthetic approach that was used to prepare the thionucleosides is outlined in the sequence (V → XIII). Thus treatment of thymidine (Va) with 1.2 moles of *p*-tolylsulfonyl chloride in pyridine at 0° gave 63% of the 5′-tosylate (VIa). That VIa was the desired 5′-tosylate of thymidine and not the isomeric 3′-tosylate was demonstrated by the preparation of the same compound by the unequivocal five-step synthesis described by Michelson and Todd[3] for the preparation of VIa.

Treatment of the tosylate (VIa) with acetic anhydride in pyridine gave the 3′-O-acetate (VIIa) as a sirup. Displacement of the tosyl group was accomplished by means of potassium thioacetate in acetone at room temperature affording 3′,5′-O,S-diacetyl-5′-thiothymidine (Xa) as a crystalline compound in 45% yield. In a similar fashion, VIa was treated with benzoyl chloride and propionyl chloride to give the benzoate (IXa) and propionate (VIIIa), respectively. Subsequent displacement of the appropriate

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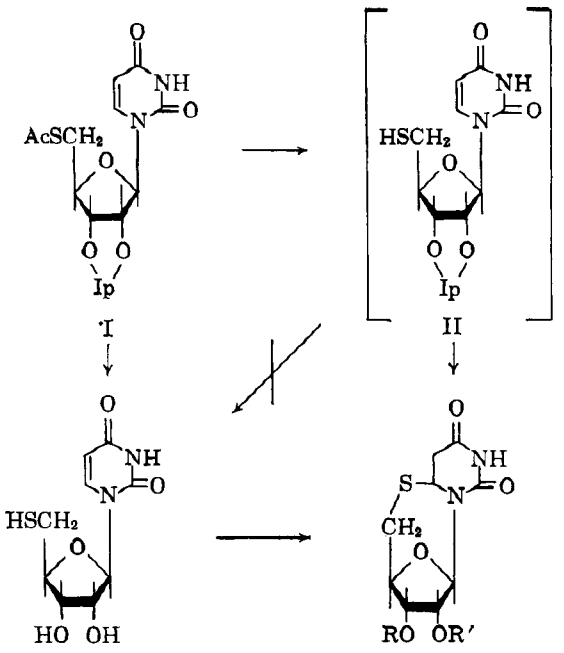
(2) (a) B. Bannister and F. Kagan, *J. Am. Chem. Soc.*, **82**, 3363 (1960); (b) R. W. Chambers and V. Kurkov, *ibid.*, **85**, 2160 (1963).

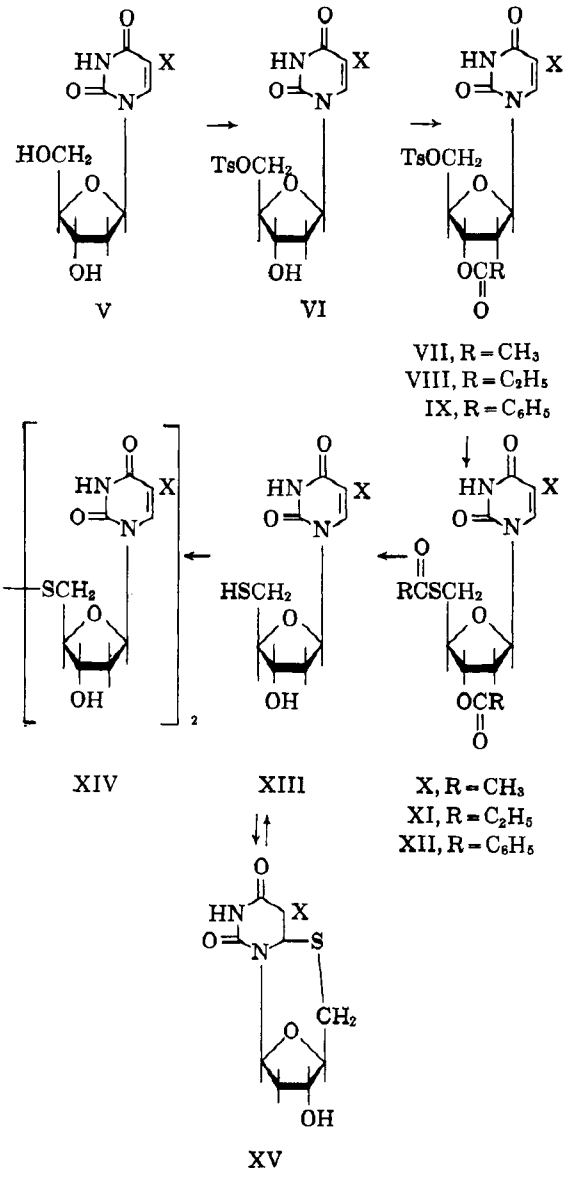
(3) A. M. Michelson and A. R. Todd, *J. Chem. Soc.*, 816 (1955).

SCHEME I



SCHEME II

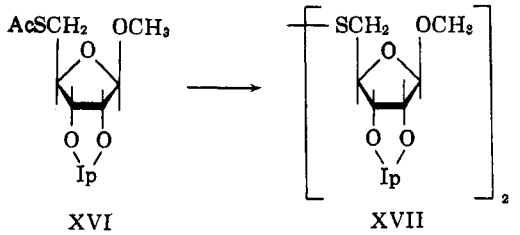
(a series, X = CH_3; b series, X = F)



acyl tosylate with potassium thiolbenzoate and potassium thiolpropionate gave the dibenzoate (XIIa) and the dipropionate (XIa). (See Scheme II.)

Similarly, 2′-deoxy-5-fluorouridine (Vb) was tosylated to give 2′-deoxy-5-fluoro-5′-O-(*p*-tolylsulfonyl)-uridine (VIb) in 58% yield. Treatment of the tosylate (VIb) with acetic anhydride in pyridine, then potassium thiolacetate gave the O,S-diacetate (Xb) and, by analogous reactions, the O,S-dibenzoate (XIIb) and the O,S-dipropionate (XIb).

The O,S-diacetate (Xa) of 5′-thiothymidine was deacetylated with methanolic ammonia. The resulting crystalline solid gave a negative nitroprusside test and showed no infrared evidence for a mercaptan group. The ultraviolet spectrum was that of a typical thymine nucleoside, however, showing that cyclization had not occurred but that oxidation to the disulfide (XIVa) had taken place. A similar oxidation to the disulfide (XVII) was observed by Clayton and Hughes[4] during the deacetylation with methanolic ammonia of methyl 5-S-acetyl-2,3-O-isopropylidene-5-thio-β-D-ribofuranoside (XVI).



Acid-catalyzed methanolysis of the O,S-diacetate (Xa) under a nitrogen atmosphere gave 5′-thiothymidine (XIIIa) as a crystalline solid that contained one thiol group by iodometric titration and which showed the typical thymidine ultraviolet absorption spectrum.

By the same procedure, treatment of the O,S-diacetate (Xb) with methanolic hydrogen chloride gave the unblocked nucleoside (XIIIb).

The isolation of the two nucleosides (XIIIa and b) from an acid solution indicated that neither one had as great a tendency as (2′,3′-O-isopropylidene-5′-thioribofuranosyl)uracil (II) towards cyclonucleoside formation. It was of interest to determine whether solutions of the 5′-mercaptonucleosides (XIIIa and b) would show any evidence for cyclonucleoside formation. The disappearance of the ultraviolet absorption in the region of 260 mμ over a period of time could be regarded as indicative of cyclonucleoside formation, while a decrease in the iodine titer,[5] a measure of the thiol content, could signify either cyclonucleoside formation or formation of the disulfide.

The results of the iodine titrations and ultraviolet absorption as a function of time are given in Table I. The results for the 5′-thiothymidine are consistent with

(4) C. J. Clayton and N. A. Hughes, *Chem. Ind.* (London), 1795 (1962).

(5) S. D. Nogare in "Organic Analysis," Vol. I, J. Mitchell, Jr., I. M. Kolthoff, E. S. Proskauer, and A. Weissberger, Ed., Interscience Publishers, Inc., New York, N. Y., 1953, p. 343.

disulfide formation at pH 13. In acid and neutral solution, there was no evidence for cyclonucleoside or disulfide formation. For the 5-fluoronucleoside (XIIIb) again, at pH 13, disulfide (XIVb) formation occurred relatively rapidly. At pH 7, the ultraviolet spectrum and iodine titer decreased more or less coincidentally over the first 24 hr., then the ultraviolet absorption increased in intensity, while the iodine titer continued to decline. At pH 13 it is clear that oxidation to the disulfide XIVb is rapid. The data at pH 7 can be rationalized by assuming a fairly slow attainment of equilibrium between XIIIb and the cyclonucleoside XVb. At this pH, XIIIb is also in equilibrium with its anion which is oxidized to the disulfide (XIVb) irreversibly. Thus the initial decrease of the ultraviolet absorption intensity reflects cyclonucleoside formation while the gradual restoration of the ultraviolet peak is the result of eventual disulfide formation. The somewhat low iodine titer recorded at pH 1 was probably the result of contamination of that sample by disulfide.

TABLE I

TIME STUDIES OF IODINE TITRATIONS AND ULTRAVIOLET ABSORPTION OF THIOLNUCLEOSIDES

		XIIIa		XIIIb	
Time, hr.	pH	Iodine titration[a]	Ultraviolet absorption[b]	Iodine titration[a]	Ultraviolet absorption[b]
0	1	100	100	82	100
	7	101	100	73	74
	13	96	100	101	100
24	1	101	100	83	95
	7	101	100	52	40
	13	8	100	6	100
96	1	110		87	96
	7	108		24	67
	13				100
300	1				100
	7				96
	13				100

[a] % of 5′-thiol according to iodometric titration. [b] Per cent of pyrimidine nucleoside according to ultraviolet spectrum.

In view of the success of acid-catalyzed methanolysis in obtaining the free 5′-thionucleosides of thymine and 5′-fluorouracil (XIIIa and XIIIb), it was of interest to investigate the acid-catalyzed methanolysis of 5′-S-acetyl-2′,3′-O-isopropylidene-5′-thiouridine (I)[2a] as a means of preparing 5′-thiouridine (III).[6] Treatment of I with methanolic hydrogen chloride gave an amorphous solid which analyzed satisfactorily for a solvate of 5′-thiouridine (III). The thiol titration at pH 7 suggested that the amorphous solid contained 80% of the nucleoside (III), the remainder, presumably, being the cyclonucleoside (IVb).

The ultraviolet spectrum of freshly prepared solutions of III at pH 1 and 7 were identical and gave an extinction coefficient of 76% of that reported at pH 3 for the disulfide of II.[2b] At pH 13, the extinction coefficient of III was 97% of that reported at pH 12 for the disulfide of II. After 5 days, there was no change in the ultraviolet spectrum of III at pH 1. At pH 7, however, after 13 days the extinction coefficient had increased to 87% of that reported for the disulfide of II at pH 3.[2b] At pH 13, again there was no change in the absorption maximum over a period of time. These results are consistent with the interpretation that at pH 1–7, a solution of III contains approximately 20% of the cyclonucleoside (IVb). This equilibrium mixture is stable at pH 1. At pH 7, III is slowly converted to disulfide thus shifting the equilibrium of III and IVb in favor of III which increases the amount of ultraviolet absorption of the solution. At pH 13, the solution contains the anion of III which is oxidized to the disulfide, a process which would not be expected to have any appreciable effect on the ultraviolet spectrum. This contrasts with the behavior of 2′,3′-O-isopropylidene-5′-thiouridine (II) which cyclized completely to the cyclonucleoside (IVa)[2b] at pH 3–7. Under strongly basic conditions, the cyclonucleoside IVa opened, and oxidation to the disulfide apparently occurred. The difference between 5′-thiouridine (III) and its 2′,3′-acetonide (II) towards cyclonucleoside formation must lie in the relative position of the 5′-mercapto group with respect to the uracil double bond in the two compounds (II and III). Evidently the isopropylidene group of II forces the furanose sugar ring into a conformation that favors the proximity of the thiol group to the uracil double bond.

(6) The synthesis of 5′-thiouridine (III) from 5′-O-*p*-(tolylsulfonyl)-uridine is claimed by A. M. Michelson [*J. Chem. Soc.*, 979 (1962)]. However, no experimental details or physical properties of III were given.

Experimental[7]

5′-O-(*p*-Tolylsulfonyl)thymidine (VIa).—A solution of 10.0 g. (41.5 mmoles) of thymidine (Va) in 50 ml. of dry pyridine was cooled to 0°, and 9.80 g. (51.5 mmoles) of *p*-tolylsulfonyl chloride in 50 ml. of dry pyridine was added dropwise with stirring. The reaction mixture was stored at 0° for 16 hr., then was decomposed with a small piece of ice and poured into 250 ml. of ice–water. The aqueous mixture was extracted with three 200-ml. portions of chloroform. The chloroform layers were combined, extracted with three 200-ml. portions of saturated aqueous sodium bicarbonate, then washed with 200 ml. of water and dried over magnesium sulfate. Evaporation of the chloroform to dryness *in vacuo* gave the crude product (VIa) as a semicrystalline solid.

Recrystallization from 250 ml. of 95% ethanol gave 10.29 g. (63%) of white crystals, m.p. 168–169° dec. Michelson and Todd[3] reported m.p. 172° dec. A mixture melting point with material prepared by their method gave no depression. The infrared spectra of the product prepared by both routes were identical.

3′,5′-O,S-Diacetyl-5′-thiothymidine (Xa).—To a solution of 5.0 g. (12.6 mmoles) of 5′-O-(*p*-tolylsulfonyl)thymidine (VIa) in 65 ml. of dry pyridine was added 5.0 ml. (53.0 mmoles) of acetic anhydride. The reaction was stored at room temperature for 16 hr., then was decomposed by the addition of 10 ml. of methanol. The solution was evaporated to dryness *in vacuo* to give 6.1 g. of 3′-O-acetyl-5′-O-(*p*-tolylsulfonyl)thymidine (VIIa) as an oily residue. The last traces of acetic acid were removed by the addition and evaporation *in vacuo* of 25 ml. of toluene.

A suspension of 4.5 g. (10.4 mmoles) of the residual gum and 4.79 g. (41.1 mmoles) of potassium thiolacetate[2] in 82 ml. of acetone was stirred at room temperature under a nitrogen atmosphere for 5 hr. The reaction mixture was cooled to 0°, then filtered, and the filter cake was washed with 50 ml. of acetone. The combined filtrate and washings were evaporated to dryness *in vacuo*. The residue was crystallized from 75 ml. of water to give 1.60 g. (45%) of white crystals, m.p. 142–143°.

Two more recrystallizations gave the analytical sample, m.p. 144–145°, $[\alpha]^{22}D$ +9.8° (1% in chloroform), $\lambda_{max}^{pH\ 1.7}$ 264 mμ (ε

(7) Melting points are corrected and were obtained with the Fisher-Johns apparatus. Paper chromatograms were run with water-saturated butyl alcohol (solvent A), 5% aqueous disodium phosphate (solvent B), and butanol-acetic acid–water (4:1:5, solvent C) by the descending technique on Whatman No. 1 paper. The spots were located by visual examination with an ultraviolet lamp. Adenine was used as a standard, and spot locations were expressed as R_{Ad} 1.00.

10,700), $\lambda_{max}^{pH\ 13}$ 267 mμ (ε 8570). The product was homogeneous on paper chromatography in solvents A, B, and C and had R_{Ad} values of 2.86, 2.08, and 1.80, respectively.

Anal. Calcd. for $C_{14}H_{18}N_2O_6S$: C, 49.1; H, 5.30; N, 8.18; S, 9.37. Found: C, 49.4; H, 5.40; N, 8.06; S, 9.55.

3′-O-Benzoyl-5′-O-(*p*-tolylsulfonyl)thymidine (IXa).—A solution of 3.73 g. (9.4 mmoles) of 5′-O-(*p*-tolylsulfonyl)thymidine (VIa) in 22 ml. of dry pyridine was cooled to 0°, then 1.38 ml. (11.7 mmoles) of benzoyl chloride was added. The reaction was stored at 0° for 16 hr. then poured slowly with stirring into 280 ml. of ice–water. The solid which separated was collected and washed with water to yield 4.59 g. (98%) of product, m.p. 171–174° dec., which was homogeneous on paper chromatography in solvents A and B and was of satisfactory purity for the next step.

The analytical sample from a previous reaction had m.p. 174–175° dec., $[\alpha]^{26}D$ −15° (1% in chloroform, $\lambda_{max}^{pH\ 1}$ 266 mμ (ε 6740), $\lambda_{max}^{pH\ 7}$ 263 mμ (ε 11,500), $\lambda_{max}^{pH\ 13}$ 266 mμ (ε 8560).

Anal. Calcd. for $C_{24}H_{24}N_2O_8S$: C, 57.6; H, 4.83; N, 5.60; S, 6.41. Found: C, 57.8; H, 4.67; N, 5.53; S, 6.35.

3′,5′-O,S-Dibenzoyl-5′-thiothymidine (XIIa).—A mixture of 4.6 g. (9.2 mmoles) of 3′-O-benzoyl-5′-O-(*p*-tolylsulfonyl)thymidine (IXa) and 6.7 g. (38 mmoles) of potassium thiolbenzoate[8] in 120 ml. of acetone was stirred at room temperature under a nitrogen atmosphere for 5 hr. The mixture was cooled to 0° and then filtered. The filter cake was washed with acetone, and the filtrate and washings were evaporated to dryness *in vacuo*. The residue was partitioned between 50 ml. each of chloroform and water. The chloroform layer was dried over magnesium sulfate, then evaporated to dryness *in vacuo* to give 4.2 g. of residue. Recrystallization from 500 ml. of 2-propanol gave 3.0 g. (71%) of white crystals, m.p. 196–197°.

The analytical sample had m.p. 199–200°, $[\alpha]^{26}D$ −2° (1% in chloroform), $\lambda_{max}^{pH\ 1}$ 268.5 mμ (ε 14,300), $\lambda_{max}^{pH\ 7}$ 268.5 mμ (ε 15,600), $\lambda_{max}^{pH\ 13}$ 265 mμ (ε 9650).

Anal. Calcd. for $C_{24}H_{22}N_2O_6S$: C, 61.8; H, 4.75; N, 6.01; S, 6.87. Found: C, 61.9; H, 4.36; N, 5.90; S, 7.06.

3′,5′-O,S-Dipropionyl-5′-thiothymidine (XIa).—A solution of 5.0 g. (12.7 mmoles) of 5′-O-(*p*-tolylsulfonyl)thymidine (VIa) in 25 ml. of dry pyridine was cooled to 0°, then 1.40 ml. (16.1 mmoles) of freshly distilled propionyl chloride was added dropwise with stirring and continued cooling. After the addition was complete, the reaction was left at room temperature for 20 hr. then was poured with stirring into 100 ml. of ice–water. The aqueous mixture was extracted with three 30-ml. portions of chloroform. The combined chloroform extracts were washed with 50 ml. of saturated aqueous sodium bicarbonate and 50 ml. of water, then dried over magnesium sulfate and evaporated to dryness *in vacuo*. The last traces of pyridine were removed by the addition and removal *in vacuo* of 10 ml. of toluene. The sirupy 3′-O-propionyl-5′-O-(*p*-tolylsulfonyl)thymidine (VIIIa) weighed 5.64 g. (99%) and was homogeneous on paper chromatography in solvents A and C with R_{Ad} values of 2.84 and 1.73, respectively.

Anal. Calcd. for $C_{20}H_{24}N_2O_8S$: N, 6.19. Found: N, 6.47.

Potassium thiolpropionate was prepared from thiopropionic acid[9] by the method used for the preparation of potassium thiolbenzoate.[8]

A mixture of 2.93 g. (22.9 mmoles) of potassium thiolpropionate and 2.50 g. (5.52 mmoles) of 3′-O-propionyl-5′-O-(*p*-tolylsulfonyl)thymidine (VIIIa) in 70 ml. of acetone was stirred at room temperature under a nitrogen atmosphere for 5 hr. The reaction was worked up in the manner described for the preparation of the dibenzoate (XIIa) to give 1.94 g. (95%) of an amorphous solid, $[\alpha]^{25}D$ +8.6° (1% in chloroform), $\lambda_{max}^{pH\ 1}$ 263.5 mμ (ε 10,700), $\lambda_{max}^{pH\ 7}$ 264.5 mμ (ε 10,200), $\lambda_{max}^{pH\ 13}$ 264.5 mμ (ε 8640). The material was homogeneous on paper chromatography in solvents A and C and had R_{Ad} values of 2.61 and 1.66, respectively.

Anal. Calcd. for $C_{16}H_{22}N_2O_6S$: C, 51.9; H, 5.99; N, 7.56; S, 8.66. Found: C, 51.9; H, 5.91; N, 7.66; S, 8.76.

5′-Thiothymidine (XIIIa).—A suspension of 6.20 g. of 3′,5′-O,S-diacetyl-5′-thiothymidine (Xa) in 62 ml. of 1 *N* methanolic hydrogen chloride was heated at 45° for 2 hr. under a nitrogen atmosphere. The small amount of insoluble material was removed by filtration and the filtrate was evaporated to dryness *in vacuo*. The residue was dissolved in 100 ml. of hot ethanol containing 1 ml. of 2-mercaptoethanol to minimize disulfide formation. The volume was reduced to 50 ml. and the solution was cooled, then filtered, and washed with cold ethanol to give 2.84 g. (61%) of white crystals, m.p. 182–194°.

The analytical sample from an earlier run had m.p. 153–154° dec., $[\alpha]^{26}D$ +47° (1% in 2-methoxyethanol), $\lambda_{max}^{pH\ 1}$ 261 mμ (ε 10,300), $\lambda_{max}^{pH\ 7}$ 261 mμ (ε 9700). The product was homogeneous on paper chromatography in solvents A and B and had R_{ad} values of 2.91 and 1.92, respectively.

Anal. Calcd. for $C_{16}H_{17}FN_2O_7S$: C, 48.0; H, 4.28; N, 7.00; S, 8.01. Found: C, 48.3; H, 4.54; N, 6.92; S, 8.28.

2′-Deoxy-3′,5′-O,S-diacetyl-5-fluoro-5′-thiouridine (Xb) was prepared from 3.5 g. of 2′-deoxy-5-fluoro-5′-O-(*p*-tolylsulfonyl)-uridine (VIb) by the procedure described for the preparation of 3′,5′-O,S-diacetyl-5′-thiothymidine (Xa) to give a 71% yield of crude product as a gummy white solid. Trituration with 60 ml. of ether gave 1.14 g. (37%) of white crystals, m.p. 87–89°, $\lambda_{max}^{pH\ 1}$ 267.5 mμ (ε 9100), $\lambda_{max}^{pH\ 7}$ 267.5 mμ (ε 8360), $\lambda_{max}^{pH\ 13}$ 268 mμ (ε 7310).

Anal. Calcd. for $C_{13}H_{15}FN_2O_6S$: C, 45.1; H, 4.37; N, 8.09; S, 9.26; F, 5.49. Found: C, 45.1; H, 4.59; N, 8.16; S, 9.08; F, 5.69.

2′-Deoxy-3′,5′-O,S-dibenzoyl-5-fluoro-5′-thiouridine (XIIb).—A solution of 5.76 g. (14.1 mmoles) of 2′-deoxy-5-fluoro-5′-O-(*p*-tolylsulfonyl)uridine (VIb) in 35 ml. of dry pyridine was cooled to 0°, then 2.1 ml. (18 mmoles) of benzoyl chloride was added dropwise with stirring. The reaction mixture was stored at 0° for 18 hr., then poured slowly with stirring into 200 ml. of ice–water. The gummy precipitate which separated was triturated with 100 ml. of acetone then 225 ml. of acetonitrile to give 4.05 g. of crude 3′-O-benzoyl-2′-deoxy-5-fluoro-5′-O-(*p*-tolylsulfonyl)-uridine (IXb) which was free of hydroxyl in the infrared and was satisfactory for further reaction.

The material could be recrystallized from acetonitrile to give material with m.p. 199–200° after three recrystallizations, $[\alpha]^{26.5}D$ +2° (1% in chloroform).

Anal. Calcd. for $C_{23}H_{21}FN_2O_8S$: C, 54.8; H, 4.20; F, 3.77; N, 5.55; S, 6.36. Found: C, 55.6; H, 4.52; F, 4.00; N, 5.93; S, 6.62.

Displacement of the *p*-tolylsulfonyl group of 3.85 g. of crude IXb by potassium thiolbenzoate in the manner described for the preparation of 3′,5′-O,S-dibenzoyl-5′-thiothymidine (XIIa) gave 3.56 g. (99%) of crude product that was free of infrared sulfonate absorption at 8.5 and 12.3 μ. Recrystallization from 400 ml. of methanol gave 1.71 g. of product, m.p. 191–192°.

The analytical sample prepared from a previous reaction had m.p. 192–193°, $[\alpha]^{26}D$ −2° (1% in chloroform), $\lambda_{max}^{pH\ 1}$ 268 mμ (ε 14,600), $\lambda_{max}^{pH\ 7}$ 267 mμ (ε 8020), $\lambda_{max}^{pH\ 13}$ 270 mμ (ε 10,000).

Anal. Calcd. for $C_{23}H_{19}FN_2O_8S$: C, 58.7; H, 4.07; N, 5.95; S, 6.82. Found: C, 58.8; H, 4.16; N, 5.88; S, 6.79.

2′-Deoxy-3′,5′-O,S-dipropionyl-5-fluoro-5′-thiouridine (XIb).—Treatment of 6.0 g. of 2′-deoxy-5-fluoro-5′-O-(*p*-tolylsulfonyl)-uridine (VIb) with propionyl chloride in pyridine in the manner described for the preparation of the propionate (VIIIa) gave 5.93 g. (90%) of 2′-deoxy-5-fluoro-3′-O-propionyl-5′-O-(*p*-tolylsulfonyl)uridine (VIIIb) as a tan foam that was homogeneous on paper chromatography in solvents A and C and had R_{Ad} values of 2.87 and 1.65, respectively, and $\lambda_{max}^{pH\ 1}$ 265.5 mμ (ε 9060), $\lambda_{max}^{pH\ 7}$ 265.5 mμ (ε 8580), $\lambda_{max}^{pH\ 13}$ 265.5 mμ (ε 7150).

Anal. Calcd. for $C_{19}H_{21}FN_2O_8S$: C, 50.0; H, 4.64; F, 4.16; N, 6.14; S, 7.02. Found: C, 49.9; H, 4.74; F, 4.30; N, 5.98; S, 6.74.

Treatment of 3.26 g. of the above tosylate (VIIIb) with potassium thiolpropionate in the manner described for the preparation of the O,S-dipropionate of 5′-thiothymidine (XIa) gave a 94% yield of 2′-deoxy-3′,5′-dipropionyl-5-fluoro-5′-thiouridine (XIb) as an amorphous solid, $[\alpha]^{25}D$ +22° (0.87% in chloroform), $\lambda_{max}^{pH\ 1}$ 266 mμ (ε 8550), $\lambda_{max}^{pH\ 7}$ 266 mμ (ε 7750), $\lambda_{max}^{pH\ 13}$ 266 mμ (ε 6860).

Anal. Calcd. for $C_{15}H_{19}FN_2O_6S$: C, 48.1; H, 5.12; F, 5.07; N, 7.48; S, 8.56. Found: C, 47.0; H, 5.82; F, 4.76; N, 6.78; S, 8.88.

2′-Deoxy-5-fluoro-5′-thouridine (XIIIb).—A solution of 1.0 g. of 3′,5′-O,S-diacetyl-5-fluoro-5′-thiouridine (Xb) in 27 ml. of 1 *N* methanolic hydrogen chloride was heated in a nitrogen atmosphere at 45° for 2.5 hr. A small amount of insoluble material was removed by filtration, and the residue was evaporated to dryness *in vacuo* to give 0.80 g. of crude solid which was homogeneous on paper chromatography in solvent A with R_{ad} 1.81 and was free of acetate absorption in the infrared.

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Recrystallization from 1700 ml. of diethyl ether gave 0.39 g. of white solid, m.p. 97–111°, $[\alpha]^{25}D$ +35° (0.25% in water), $\lambda_{max}^{pH\ 1}$ 268 mμ (ε 8840), $\lambda_{max}^{pH\ 7}$ 268 mμ (ε 4900), $\lambda_{max}^{pH\ 13}$ 268 mμ (ε 7260). The material traveled as one component in solvent A with R_{ad} 1.81. In solvent C, it travelled as an elongated spot with R_{ad} 1.29–1.37.

Anal. Calcd. for $C_9H_{11}FN_2O_4S$: C, 41.2; H, 4.23; F, 7.24; N, 10.7; S, 12.2. Found: C, 41.4; H, 4.30; F, 7.53; N, 10.6; S, 12.1.

5'-Thiouridine (III).—A solution of 0.47 g. of 5'-S-acetyl-2',3'-O-isopropylidene-5'-thiouridine (I)[28] in 5 ml. of 1 *N* methanolic hydrogen chloride was heated at 45° for 2.5 hr. then was filtered, and the filtrate was evaporated to dryness *in vacuo* to give a sirup which gave a strong positive nitroprusside test. The sirup was triturated several times with absolute ether to afford 0.366 g. (100%) of an amorphous solid, m.p. 109–111°, $\lambda_{max}^{pH\ 1,\ 7}$ 262 mμ (ε 7320), $\lambda_{max}^{pH\ 13}$ 262 mμ (ε 6500). The ultraviolet spectra at pH 1 and 7 indicated that approximately 25% of III had cyclized to the cyclonucleoside (IVb), whereas the spectrum at pH 13 appeared to be normal. The paper chromatograms showed major spots at R_{ad} 0.19 and 0.91 in solvents A and C, respectively. Both chromatograms showed traces of contaminants.

Anal. Calcd. for $C_9H_{12}N_2O_5S \cdot 0.5\ (C_2H_5)_2O$: C, 44.4; H, 5.76; N, 9.42; S, 10.8; SH, 11.1. Found: C, 44.2; H, 5.40; N, 9.39; S, 10.6; SH, 8.8.

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Nucleosides. XVIII. Synthesis of 2'-Fluorothymidine, 2'-Fluorodeoxyuridine, and Other 2'-Halogeno-2'-Deoxy Nucleosides[1,2]

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The reaction of 2,2'-anhydro nucleosides with anhydrous hydrogen halides gave 2'-halogeno-2'-deoxy nucleosides in good yields. By this reaction the 2'-fluoro, 2'-chloro, and 2'-bromo analogs of deoxyuridine [VIIa(F), VIIa(Cl), VIIa(Br)] and of thymidine [VIIb(F), VIIb(Cl), VIIb(Br)] were synthesized. The 2'-fluoro analog of 5-fluorodeoxyuridine [VIIc(F)] was prepared. Comparison was made of the stability of the 2'-halogeno-deoxyuridine derivatives in alkali, acid, and upon heating.

The unique role of the substituent (hydrogen or hydroxyl) on the 2'-carbon atom of nucleic acids as the distinguishing feature between deoxyribonucleic acids (DNA) and ribonucleic acids (RNA) prompted an investigation of the biological properties of nucleosides containing substituents other than hydrogen or hydroxyl at this position. Accordingly, the synthesis of 2'-halogeno-2'-deoxypyrimidine nucleosides, as compounds of potential biological interest, was undertaken. Of particular interest were the 2'-fluoro-2'-deoxy nucleosides, 2'-fluorothymidine [VIIb(F)] and 2'-fluorodeoxyuridine [VIIa(F)] (see Scheme I), analogs of thymidine and 2'-deoxyuridine, respectively. It is noteworthy that among the most active known antagonists of nucleic acid biosynthesis are 5-fluorodeoxyuridine[3] and 5-trifluoromethyldeoxyuridine,[4] both possessing fluorine in place of hydrogen in the pyrimidine moiety of naturally occurring deoxy nucleosides. VIIb(F) and VIIa(F), on the other hand, possess fluorine substituted for hydrogen in the sugar moiety of naturally occurring deoxy nucleotides.

The possibility of using 2,2'-anhydro nucleosides as starting materials for the synthesis of 2'-halogeno deoxy nucleosides was investigated.[5] Such an approach was supported by the work of Brown and co-workers.[12] These authors found that 2,2'-anhydro-1-(5'-*O*-acetyl-β-D-arabinofuranosyl)uracil was an intermediate in the synthesis of 5'-*O*-acetyl-2'-iododeoxyuridine in a reaction of the 2'-tosyloxy nucleoside with sodium iodide in acetonylacetone.[13] When the 2,2'-anhydro intermediate was used under the same conditions (sodium iodide in acetonylacetone at 100°) no reaction occurred. As pointed out by Fox and Miller,[14] the successful conversion of the 2'-tosyloxy derivative to its 2'-iodo analog by these workers was probably due to the presence of a small amount of toluenesulfonic acid liberated in the formation of the 2,2'-anhydro intermediate. The acid then served to catalyze the cleavage of the anhydro bridge by iodide ion. Brown, *et al.*,[13] did convert the anhydro nucleoside to the 2'-iodo derivative in small yield only after the addition of a small amount of acetic acid.

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