triplet, J = 4 c./sec.) 6.3 (2H, singlet), 7.05 (2H, doublet, J = 4 c./sec.), 7.1 (3H, singlet), 7.55 τ (3H, singlet).

Anal. Calcd. for $C_{15}H_{17}NO_3S$: C, 61.85; H, 5.88; N, 4.81; S, 11.00. Found: C, 61.88; H, 5.97; N, 4.87; S, 10.91.

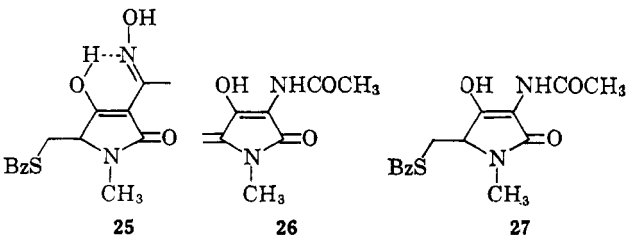
Oxime 25 was prepared from 29.1 g. (0.1 mole) of **14** by the technique described previously. However, the product did not separate from the reaction mixture. Evaporation of the ethanol left an oil–water mixture which was extracted with benzene. Removal of the solvent from the dried organic phase gave a sirup which upon slow crystallization from benzene yielded colorless crystals (27 g., 88%), m.p. 60–62°, blue color reaction with ferric chloride; $\nu_{max}^{CHCl_3}$ 3400–2500, 1670, 1610, 1580 cm.$^{-1}$; λ_{max}^{EtOH} 248, 311 mμ (ϵ 6350, 14,300).

Anal. Calcd. for $C_{15}H_{18}N_2O_3S$: C, 58.81; H, 5.92; N, 9.15; S, 10.45. Found: C, 59.20; H, 6.01; N, 8.96; S, 10.31.

1-Methyl-3-acetyl-4-hydroxy-5-methylene-3-pyrrolin-2-one (15).—When the cyclization of the N-acetoacetyl derivative **13** was performed at 80° for 3 hr., a yellow sirup was obtained which could not be induced to crystallize. Sublimation under high vacuum produced colorless plates of the diene lactam **15**, 15%, m.p. 80°; ν_{max}^{KBr} 1710, 1672, 1630, 1280, 1180, 940, 860 cm.$^{-1}$; λ_{max}^{EtOH} 228, 267 mμ (ϵ 8350, 23,400); n.m.r. ($CDCl_3$) −2.5 (1H, broad singlet), 4.66 (1H, triplet $J \sim 1$ c./sec.), 5.3 (1H, multiplet), 6.90 (3H, doublet $J \sim 1$ c./sec.), 7.45 τ (3H, narrowly spaced doublet).

Beckmann Rearrangement of Oxime 25.—Sodium hydroxide (0.24 g., 6 mmoles) in water (24 ml.) was added to a stirred solution of oxime **25** (0.61 g., 2 mmoles) and tosyl chloride (0.38 g., 2 mmoles) in 20 ml. of acetone at 50° during 20 min. After 12 hr. at 65° the solution was cooled, acidified with dilute hydrochloric acid, and concentrated. Extraction of the residue with benzene and removal of the solvent from the dried extract left a mixture of an oil and a small amount of light yellow crystals. Ether was added to dissolve the oil and the crystals were collected. In this manner light yellow cubes (0.066 g., 19%), m.p. *ca.* 190° (with sublimation), were obtained. Recrystallization from ethanol afforded an analytical sample of the substituted acetamide **26**. The pure compound gave a faint blue color reaction with ferric chloride; ν_{max}^{KBr} 3400–2600, 1690, 1670, 1640, 1540, 880, 840 cm.$^{-1}$; λ_{max}^{EtOH} 268, 342 mμ (ϵ 20,600, 1740); n.m.r. ($CDCl_3$) −1.00 (1H, singlet), 4.83 (1H, doublet, J = 2 c./sec.), 5.18 (1H, doublet, J = 2 c./sec.), 6.92 (3H, singlet), 7.75 τ (3H, singlet).

Anal. Calcd. for $C_8H_{10}N_2O_3$: C, 52.74; H, 5.53; N, 15.38. Found: C, 52.96; H, 5.62; N, 15.72.



Slow evaporation of the ethereal filtrate left an oil which solidified partially after several days. Repeated trituration with an ether–cyclohexane mixture resulted in the separation of a solid which was recrystallized from ethanol to yield colorless needles of **27** (0.091 g., 13%), m.p. 124°. Several recrystallizations raised the melting point to 154°; $\nu_{max}^{CHCl_3}$ 3400, 3200, 3000, 1690–1660, 1630, 1540, 700 cm.$^{-1}$; λ_{max}^{EtOH} 210, 270 mμ (ϵ 20,000, 5400); blue color with ferric chloride; n.m.r. ($CDCl_3$) −1.66 (1H, singlet), 1.33 (1H, broad), 2.7 (5H, singlet), 6.05 (1H, triplet, J = 4 c./sec.), 6.28 (2H, singlet), 7.1 (2H, doublet, J = 4 c./sec.), 7.16 (3H, singlet), 7.8 τ (3H, singlet).

Anal. Calcd. for $C_{15}H_{18}N_2O_3S$: C, 58.81; H, 5.92; N, 9.15; S, 10.45. Found: C, 58.84; H, 6.05; N, 9.13; S, 10.48.

Acknowledgments.—We are indebted to the National Institutes of Health (GM 09686-03) and to Chas. Pfizer and Co. for financial support.

[CONTRIBUTION FROM LIFE SCIENCES RESEARCH, STANFORD RESEARCH INSTITUTE, MENLO PARK, CALIFORNIA]

Synthesis of 4-Thio-D- and -L-ribofuranose and the Corresponding Adenine Nucleosides[1,2]

BY ELMER J. REIST, DONALD E. GUEFFROY, AND LEON GOODMAN

RECEIVED AUGUST 14, 1964

The synthesis of 4-thio-D-ribose derivatives from L-lyxose is described. Methyl 2,3-O-isopropylidene-4-(*p*-tolylsulfonyl)-α-L-lyxopyranoside (Xa) when treated with sodium thiolbenzoate in N,N-dimethylformamide gave methyl 4-S-benzoyl-2,3-O-isopropylidene-4-thio-β-D-ribopyranoside (XIa). Deacetonation of this product followed by acetolysis gave 1,2,3,5-tetra-O-acetyl-4-thio-D-ribofuranose (XIVa) which was converted directly to 4′-thio-D-adenosine (XXVIIIa). In a similar fashion 1,2,3,5-tetra-O-acetyl-4-thio-L-ribofuranose (XIVb) and 4′-thio-L-adenosine (XXVIIIb) were prepared from D-lyxose. Deacetylation of the XIVb gave 4-thio-L-ribofuranose (IVb) as a sirup. Spectroscopic evidence indicated that IVb exists primarily in the thiofuranose form. Reactions such as acylation or glycoside formation also occur to give the thiofuranose derivatives. The sulfur atom of compound IVb easily reacts as a thiol, however, as indicated by easy disulfide formation and rapid iodine uptake, thus indicating a facile equilibrium between furanose (IV) and pyranose (VI) forms, a situation quite different from that reported for 5-thio-D-ribose.

There has been a great deal of activity in recent years in the synthesis of 5-thio sugars in which the pyranose ring oxygen has been replaced by sulfur. Thus, 5-thio-D-xylose,[3] 5-thio-D-ribose,[4] and 5-thio-D-glucose[5] among others have been synthesized. In all cases, the sulfur atom assumed the ring position to give a thiopyranose (III) configuration rather than the isomeric furanose (I) involving oxygen ring closure. A few 6-thiohexoses have been prepared[6]; however, no information was given concerning the ability of the sulfur atom to form a seven-membered ring.

The widespread occurrence in biological systems of D-ribose in its furanose form and the knowledge that substitution of sulfur for oxygen in compounds of biological importance has resulted in analogs of chemotherapeutic value made it of interest to investigate the synthesis of 4-thio-D-ribose and its derivatives.

It seemed reasonable to expect that the driving force which caused sulfur to assume the ring position of a thiopyranose might carry through to the 4-thio sugars

(1) This work was carried out under the auspices of the Cancer Chemotherapy National Service Center, National Cancer Institute, National Institutes of Health, Public Health Service, Contract No. PH43-64-500. The opinions expressed in this paper are those of the authors and not necessarily those of the Cancer Chemotherapy National Service Center.

(2) Certain portions of this work have been reported previously. See E. J. Reist, D. E. Gueffroy, and L. Goodman, *J. Am. Chem. Soc.*, **85**, 3715 (1963).

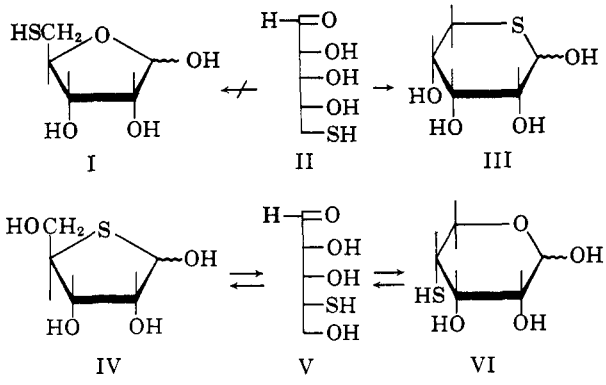
(3) (a) J. C. P. Schwarz and K. C. Yule, *Proc. Chem. Soc.*, 417 (1961); (b) T. J. Adley and L. N. Owen, *ibid.*, 418 (1961); (c) R. L. Whistler, M. S. Feather, and D. L. Ingles, *J. Am. Chem. Soc.*, **84**, 122 (1962).

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

(5) M. S. Feather and R. L. Whistler, *Tetrahedron Letters*, 667 (1962).

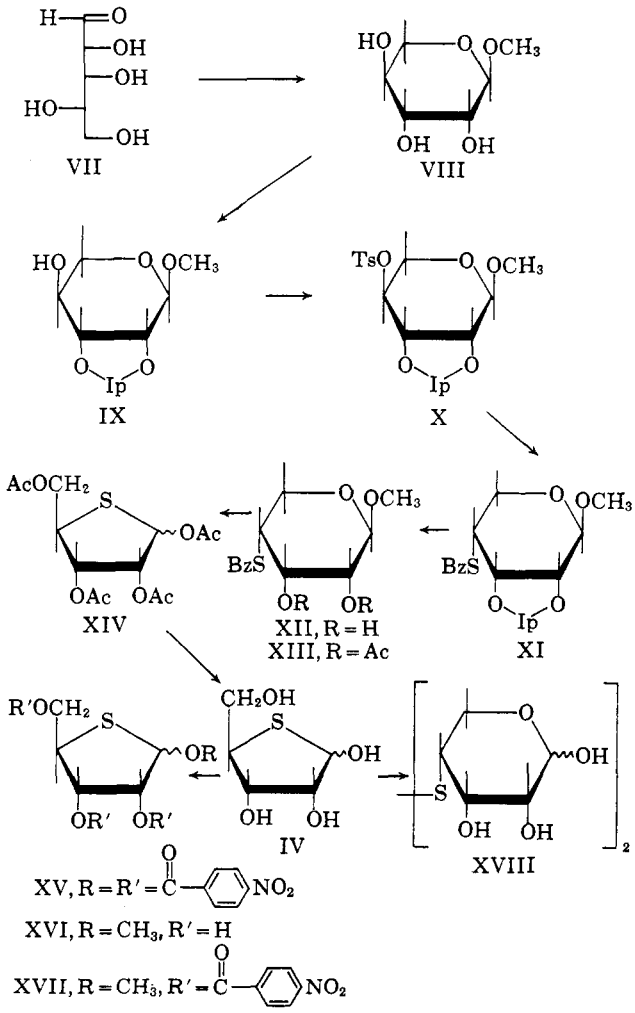
(6) (a) D. Horton and W. N. Turner, *Chem. Ind.* (London), 76 (1964); (b) M. Akagi, S. Tejima, and M. Haga, *Chem. Pharm. Bull.*, **10**, 562 (1962).

and favor the thiofuranose structure IV over the 4-thio-O-pyranose VI. Such a thiofuranose (IV) might



be converted to biologically interesting analogs such as 4′-thioadenosine (XXVIII). The synthesis of 4-thio-D-ribose derivatives from L-lyxose[7] (VIIa) and the subsequent conversion to 4′-thioadenosine is the subject of this paper.

The synthesis of methyl 2,3-O-isopropylidene-4-O-(*p*-



a series = L-lyxose → 4-thio-D-ribose
b series = D-lyxose → 4-thio-L-ribose

tolylsulfonyl)-α-L-lyxopyranoside (Xa) was accomplished by a modification of the procedure described by Kent and Ward[8] for the preparation of the analogous D-lyxoside Xb. Thus treatment of L-lyxose[7] with 0.5% methanolic hydrogen chloride gave a 65% yield of crystalline methyl α-L-lyxopyranoside (VIIIa). The use of 2,2-dimethoxypropane and hydrogen chloride in dioxane, an acetonation mixture used by Chladek and Smrt[9] for the acetonation of nucleosides, proved to be more convenient than the acetone–sulfuric acid reported and gave a quantitative yield of methyl 2,3-O-isopropylidene-α-L-lyxopyranoside (IXa) of satisfactory purity for subsequent tosylation to Xa. Thus methyl 2,3-O-isopropylidene-4-O-(*p*-tolylsulfonyl)-α-L-lyxopyranoside (Xa) was obtained in 53% over-all yield from L-lyxose (VIIa).

A key step in the reaction sequence involved the SN2 displacement of the tosylate of Xa by a sulfur-containing nucleophile to give a substituted 4-thio-D-ribopyranose. The use of sodium benzoate in N,N-dimethylformamide (DMF) has been very successful in displacing similar ring tosylates, so an analogous thiolbenzoate in DMF seemed a logical reagent for this reaction. Treatment of Xa with potassium thiolbenzoate in DMF at 115° for 72 hr. displaced the tosylate completely and a 63% yield of sirupy methyl 4-S-benzoyl-2,3-O-isopropylidene-4-thio-β-D-ribopyranoside (XIa) resulted. Deacetonation of XIa with 66% aqueous acetic acid gave crystalline methyl 4-S-benzoyl-4-thio-β-D-ribopyranoside (XIIa) in 42% yield.

At this time, the experimental work was shifted to the D-lyxoside series in view of the commercial availability of D-lyxose. Thus D-lyxose (VIIb) was converted to the crystalline thiolbenzoate XIIb and the chemistry of 4-thioribose was developed in the L-series.

Treatment of the crystalline thiolbenzoate XIIb with acetic anhydride in pyridine gave a product (XIIIb) which was not homogeneous on thin layer chromatography and which appeared to have suffered some loss of sulfur as indicated by elemental analysis. An acid-catalyzed acetylation proceeded smoothly, however, and the diacetate XIIIb was obtained as a homogeneous, analytically pure oil.

In order to effect a ring contraction to a 4-thiofuranose, it seemed necessary to open the pyranose ring of XIII to give 4-thioribose (IV–VI) which could conceivably be blocked to force the furanose ring form. The first step involved the acetolysis of the pyranose glycoside XIIIb to obtain the base labile acetate group at C-1. When XIIIb was subjected to the usual acetolysis conditions, a sirup was obtained which proved to be totally free of any detectable traces of S-benzoate, O-benzoate, or S-acetate according to infrared spectroscopy. This product was homogeneous on thin-layer chromatography and had analytical values in accord with those for 1,2,3,5-tetra-O-acetyl-4-thio-L-ribofuranose (XIVb). The same product could be obtained by the direct acetolysis of the unblocked thiobenzoate XIIb.

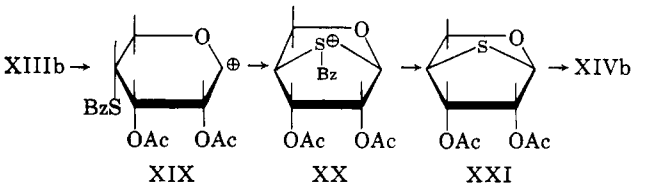
The spontaneous contraction from the pyranose ring of XIII to the furanose ring of XIV was somewhat surprising. It should be noted, however, that the reverse situation, namely a ring expansion from an O-furanose to S-pyranose, was reported by Feather and Whistler[5] during the acetolysis of 3,5,6-O,S,O-triacetyl-1,2,-O-isopropylidene-5-thio-D-glucofuranose to prepare 1,2,3,-

(7) R. L. Whistler and J. N. BeMiller in "Methods in Carbohydrate Chemistry," Vol. I, R. L. Whistler and M. L. Wolfrom, Ed., Academic Press, Inc., New York, N. Y., 1962, p. 79.

(8) P. W. Kent and P. F. V. Ward, *J. Chem. Soc.*, 416 (1953).

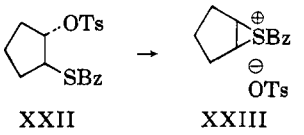
(9) S. Chladek and J. Smrt, *Collection Czech. Chem. Commun.*, **28**, 1301 (1963).

4,6-penta-O-acetyl-5-thio-D-glucopyranose. It is interesting to speculate that this ring contraction or



expansion proceeds by a mechanism such as outlined in XIIIb → XIX → XX → XXI → XIVb.[10]

Deacetylation of the tetraacetate XIVb with methanolic sodium methoxide for 3 hr. gave 4-thio-L-ribofuranose (IVb) as an analytically pure sirup which was homogeneous on thin layer chromatography and which gave a positive nitroprusside test for a thiol. Deacetylation of XIVb with methanolic sodium methoxide for 60 hr. gave 4-thio-L-ribofuranose (IVb) had a second com-

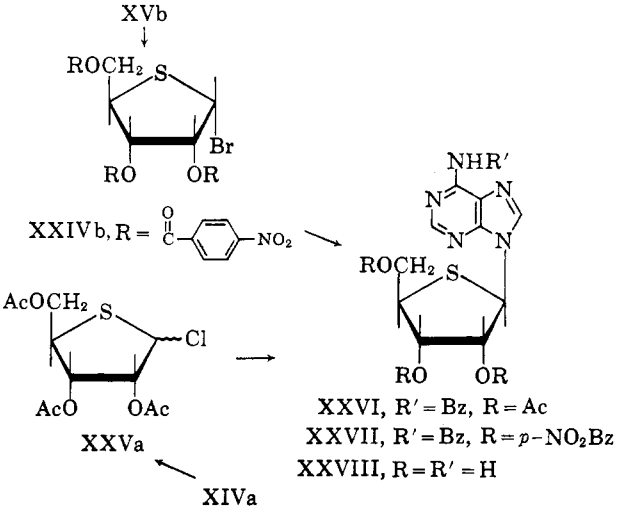


ponent which was distinguishable from IV on thin-layer chromatography. This second component could be crystallized and proved to be the disulfide XVIIIb, formed by air oxidation over the longer reaction period.

The difference between 4-thio-L-ribofuranose (IVb) and 5-thio-D-ribopyranose (III) toward thiol reagents such as aqueous iodine is striking. Clayton and Hughes[4] reported that 5-thio-D-ribopyranose (III) reacted only slowly with iodine in aqueous solution. This behavior seems typical of the 5-thio-D-pyranose sugars since Adley and Owen[3b] observed that 5-thio-D-xylopyranose required several hours and intermittent heating to take up 85% of the theoretical amount of aqueous iodine solution. On the other hand, the thiofuranose IVb reacted rapidly with aqueous iodine at room temperature to approximately the theoretical uptake for one thiol group, after which there was a relatively slow additional iodine consumption. This difference in behavior toward iodine titration together with the infrared evidence showing the absence of significant amounts of absorption at 5.75 and 3.90, μ[12] indicative of the carbonyl and free thiol, respectively, suggests that in the thiofuranose series there exists a very facile equilibrium among the three tautomers IV, V, and VI. Although at equilibrium tautomer IV predominates to the virtual exclusion of V and VI, suitable reaction conditions can easily shift the equilibrium so that reaction can occur with VI without difficulty, *e.g.*, iodine titration and air oxidation.

Acylation of 4-thio-L-ribose with *p*-nitrobenzoyl chloride in pyridine gave the crystalline tetra-*p*-nitrobenzoate XVb which showed infrared carbonyl absorption at 5.78 μ only with none near 6.0 μ, thus indicating that acylation occurred exclusively on oxygen to give the thiofuranose sugar. Reaction of IVb with methanolic hydrogen chloride gave the sirupy methyl glycoside which was assigned the furanose configuration XVIb on the basis of the absence of thiol absorption at 3.90 μ in the infrared and the failure of the glycoside to give a positive nitroprusside test. That this assignment was correct was shown by acylation of the glycoside XVIb with *p*-nitrobenzoyl chloride to give a crystalline tri-*p*-nitrobenzoate (XVIIb) which showed no evidence for S–acyl absorption in the infrared.

With two suitably blocked 4-thioribofuranose derivatives (XIVb and XVb) in hand, the preparation of a halosugar,[13] a convenient intermediate for the preparation of nucleosides, was investigated. Treatment of the



a series = 4-thio-D-ribose; b series = 4-thio-L-ribose

tetranitrobenzoate XVb with hydrogen bromide in acetic acid gave a 68% yield of a crystalline presumably anomerically pure compound which proved to be the desired 2,3,5-tri-O-(*p*-nitrobenzoyl)-4-thio-α-L-ribofuranosyl bromide (XXIVb). That this bromo sugar would react in the normal fashion of glycosyl halides was demonstrated by treating it with chloromercuri-6-benzamidopurine, to give the blocked nucleoside XXVIIb from which crystalline 4′-thio-L-adenosine (XXVIIIb) could be obtained by deacylation with methanolic sodium methoxide.

Returning to the D-series, acetolysis of methyl 4-S-benzoyl-4-thio-β-D-ribopyranoside (XIIa) with acetic anhydride and sulfuric acid in acetic acid gave a 97% yield of analytically pure 1,2,3,5-tetra-O-acetyl-4-thio-D-ribofuranose (XIVa) as a sirup which showed no evidence for S–acyl absorption in the infrared spectrum. This sirup could be crystallized from methanol to give the crystalline β-acetate. The β-configuration is assigned on the basis of the rotation of the crystals (−102°) compared with the anomeric sirup (−59°). Treatment of sirupy XIVa with hydrogen chloride in ether gave the chlorosugar XXVa as a sirup which was condensed with chloromercuri-6-benzamidopurine, then

(10) The spontaneous formation of cyclopentene S-benzoylepisulfonium *p*-toluenesulfonate (XXIII) from *trans*-2-benzoylthiocyclopentanol-*p*-toluenesulfonate (XXII) at room temperature, which was reported by Goodman, *et al.*,[11] indicates that the sulfur atom can readily form an acylsulfonium ion of the type postulated for XX.

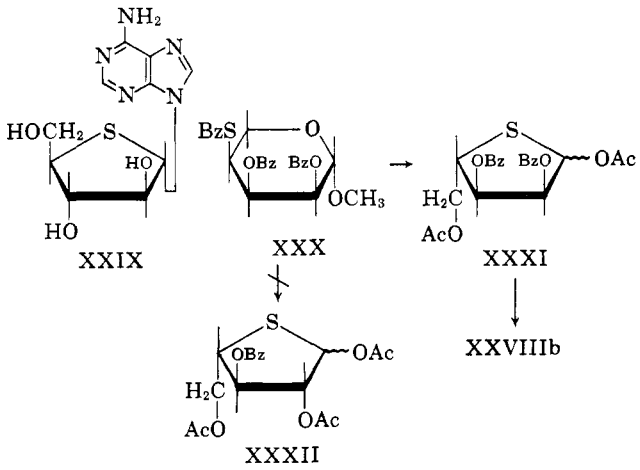
(11) L. Goodman, A. Benitez, and B. R. Baker, *J. Am. Chem. Soc.*, **80**, 1680 (1958).

(12) Debenzoylation of methyl 4-S-benzoyl-4-thio-β-L-ribopyranoside (XIIb) with methanolic sodium methoxide gave crude methyl 4-thio-β-L-ribopyranoside in which there was thiol absorption at 3.90 μ. Thus the absence of absorption at 3.90 μ in IVb is indicative of the absence of free thiol.

(13) After the completion of this portion of the work, a report by R. L. Whistler and T. Van Es, *J. Org. Chem.*, **28**, 2303 (1963), described the preparation and some reactions of 2,3,4-tri-O-acetyl-5-thio-D-xylopyranosyl bromide. This bromo sugar reacted in the normal fashion with methanol to give a methyl glycoside, although its reactivity was low, relative to the oxygen analog.

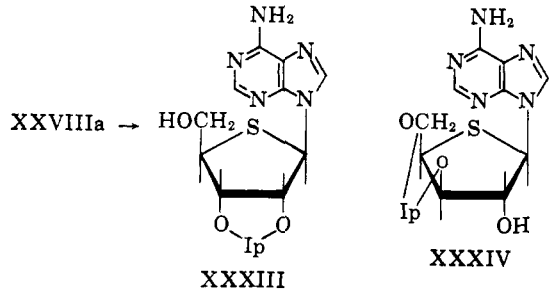
deacylated to give crystalline 4′-thio-D-adenosine (XXVIIIa) that was identical with the L-anomer in all respects except the rotation which was equal and opposite.

A recent paper by Jerkeman[14] described the epimerization of a D-mannose derivative to D-glucose during the course of an acetolysis reaction. Jerkeman suggested that a similar type of epimerization might be expected from D-ribose to its epimer D-arabinose. Presumably D-thioribose could also undergo such a rearrangement to D-thioarabinose, in which case the above nucleoside rather than being 4′-thioadenosine (XXVIII) would be 9-(4′-thio-α-D-arabinofuranosyl)adenine (XXIX). That such a rearrangement did not occur was demonstrated in the following manner: Benzoylation of the thiolbenzoate XIIb with benzoyl chloride in pyridine gave methyl 2,3,4-O,O,S-tribenzoylribopyranoside (XXX) as a sirup. Acetolysis of XXX in the usual fashion again caused ring contraction to give sirupy 1,5-di-O-acetyl-2,3-di-O-benzoyl-4-thio-L-ribose (XXXI). The significant feature in this reaction is that there was no replacement of O-benzoate by O-acetate during the acetolysis. If an epimerization of the type described by



Jerkeman[14] had occurred, this should have been accompanied by replacement of the 2-O-benzoate by acetate to give the arabinose derivative XXXII. The n.m.r. spectra of the crude acetolysis product from XXX showed a ratio of two benzoates to two acetates; hence, the product had to be the riboside XXXI. Treatment of XXXI with hydrogen chloride in ether followed by condensation with chloromercuri-6-benzamidopurine gave, after deacylation, 4′-thioadenosine (XXVIIIb) which was identical in every respect with the 4′-thioadenosine prepared from the tetraacetate XIVb.

Added confirmation that the nucleosides XXVIII were ribosides and not arabinosides was obtained by acetonation of the nucleoside with acetone and *p*-toluenesulfonic acid to give a 68% yield of the isopropylidene nucleoside XXXIII. The fact that the nucleoside XXVIIIa had two hydroxyls situated in the necessary steric relationship to permit formation of an isopropylidene derivative eliminates the D-arabinofuranose conformation from consideration, although a 3,5-O-isopropylidenexylofuranose nucleoside (XXXIV) is possible. That the nucleoside was not XXXIV was strongly suggested by the similarity in n.m.r. spectra between XXXIII and 2′,3′-O-isopropylideneadenosine, together with the dissimilarity between the n.m.r. spectra of



XXXIII and (3′,5′-O-isopropylidene-β-D-xylofuranosyl)adenine.

Experimental[15]

Methyl α-L-Lyxopyranoside (VIIIa).—A solution of 0.8 g. of L-lyxose[7] in 16 ml. of 0.5% methanolic hydrogen chloride was heated at reflux for 5 hr. At the end of this time, the solution was neutralized with silver carbonate, then filtered through a Celite pad. The filtrate was evaporated to dryness to give a sirup which crystallized from ethyl acetate affording 550 mg. (65%) of white crystals, m.p. 102–104°. The analytical sample had m.p. 104.5–106.0°, $[\alpha]^{26}D$ −49° (1% in water).

Anal. Calcd. for $C_6H_{12}O_5$: C, 43.9; H, 7.32. Found: C, 44.2; H, 7.25.

Methyl α-D-lyxopyranoside[8] (VIIIb) has m.p. 108–109°, $[\alpha]D$ +52° (0.6% in water).

Methyl 2,3-O-Isopropylidene-α-L-lyxopyranoside (IXa).—To a stirred suspension of 39.2 g. of methyl α-L-lyxopyranoside (VIIIa) in 900 ml. of 2,2-dimethoxypropane was added 10 ml. of 4 *M* hydrogen chloride in dioxane.[9] The reaction was stirred overnight at room temperature by which time solution was complete. The reaction was neutralized with solid sodium bicarbonate, then filtered through a Celite pad, and the filtrate was evaporated to dryness *in vacuo*. The resulting methyl 2,3-O-isopropylidene-α-L-lyxopyranoside (IXa) was a pale yellow oil which crystallized on standing and was of satisfactory purity for the next step. The analytical sample, obtained by recrystallization from benzene–hexane (1:1), had m.p. 51.5–52.5°, $[\alpha]^{25}D$ −47° (1% in ethanol).

Anal. Calcd. for $C_9H_{16}O_5$: C, 52.9; H, 7.90. Found: C, 53.0; H, 7.97.

Verheyden and Stoffyn[16] reported m.p. 49–52°, $[\alpha]^{25}D$ +45.6° (EtOH), for the D-analog IXb.

Methyl 2,3-O-Isopropylidene-4-O-(*p*-tolylsulfonyl)-α-L-lyxopyranoside (Xa).—A solution of 49.0 g. (0.24 mole) of crude methyl 2,3-O-isopropylidene-α-L-lyxopyranoside (IXa) in 600 ml. of dry pyridine was evaporated *in vacuo* until 500 ml. of solution remained. To this solution, chilled to 0°, was added 135 g. (0.69 mole) of *p*-tolylsulfonyl chloride, and the reaction mixture was stirred at room temperature for 4 days. The solution was added dropwise with stirring to 3 l. of saturated aqueous sodium bicarbonate solution. The solid that formed slowly crystallized and was recrystallized twice from 95% ethanol affording 68.8 g. (81%) of solid, m.p. 103.5–104.0°. The analytical sample had m.p. 104.5–105.0° and $[\alpha]^{25}D$ +11.5° (1% in chloroform).

Anal. Calcd. for $C_{16}H_{22}O_7S$: C, 53.6; H, 6.19; S, 8.95. Found: C, 53.5; H, 6.20; S, 9.14.

Kent and Ward[8] gave m.p. 96–97°, $[\alpha]^{22}$ −10.2° (*c*, 1.85 in ethanol) for the α-D isomer (Xb).

Methyl 4-S-Benzoyl-4-thio-β-D-ribopyranoside (XIIa).—A mixture of 35.0 g. (97 m*M*) of methyl 2,3-O-isopropylidene-4-O-(*p*-tolylsulfonyl)-α-L-lyxopyranoside (Xa) and 56 g. (3.2 moles) of potassium thiolbenzoate[17] in 1000 ml. of dry DMF was heated at

(14) P. Jerkeman, *Acta Chem. Scand.*, **17**, 2769 (1963).

(15) Melting points were taken on a Fisher-Johns apparatus and are corrected. Magnesium sulfate was used as the drying agent throughout. Paper chromatograms were run by the descending procedure on Whatman No. 1 paper. The spots were located by visual examination with an ultraviolet lamp. Thin layer chromatograms were run on silicic acid. The spots were located by a sulfuric acid spray. The solvent systems used were A, 1-butanol–acetic acid–water (5:2:3); B, water–saturated 1-butanol; C, 5% aqueous disodium hydrogen phosphate; D, ethyl acetate–methanol (1:1).

(16) J. P. Verheyden and P. J. Stoffyn, *Tetrahedron*, **1**, 253 (1957).

(17) T. P. Johnston and A. Gallagher, *J. Org. Chem.*, **26**, 3780 (1961).

115° for 72 hr. The reaction mixture was evaporated to dryness *in vacuo*, and the residue was partitioned between 500 ml. each of ether and water. The aqueous layer was extracted with two 100-ml. portions of ether. The combined ether layers were washed with six 500-ml. portions of water, then dried and evaporated to dryness *in vacuo*. The resulting methyl 4-S-benzoyl-2,3-O-isopropylidene-4-thio-β-D-ribopyranoside (XIa), 20.0 g. (63%), was a pale yellow oil, $\lambda_{max(\mu)}^{film}$ 6.0 (thiobenzoyl C=O); there was no tosylate absorption at 8.5 μ.

A solution of 20.0 g. of crude methyl 4-S-benzoyl-2,3-O-isopropylidene-4-thio-β-D-ribopyranoside (XIa) in 450 ml. of 66% aqueous acetic acid was heated at 50° for 3 hr., then evaporated to dryness *in vacuo* at a temperature not greater than 40°. The resulting yellow sirup was dissolved in 100 ml. of chloroform and washed with 100 ml. of water. The chloroform layer was dried and evaporated to dryness *in vacuo*. The solid residue was recrystallized from benzene, then twice from isopropyl alcohol to give 7.4 g. (42%) of white crystals, m.p. 152.0–152.5°. The analytical sample from a previous run had m.p. 148.0–149.5°, $[\alpha]^{25}D$ −70° (1% in ethanol), $\lambda_{max(\mu)}^{Nujol}$ 2.85, 3.05 (OH), 6.05 (thiolbenzoyl C=O).

Anal. Calcd. for $C_{13}H_{16}O_5S$: C, 54.9; H, 5.67; S, 11.3. Found: C, 54.9; H, 5.64; S, 11.3.

Methyl 4-S-Benzoyl-4-thio-β-L-ribopyranoside (XIIb) was made by an identical procedure from methyl 2,3-O-isopropylidene-4-O-(*p*-tolylsulfonyl)-α-D-lyxopyranoside[16] (Xb) to give the product as white crystals, m.p. 148.0–149.5°, $[\alpha]^{24}D$ +72° (1% in ethanol).

Anal. Found: C, 54.8; H, 5.66; S, 11.2.

Methyl 2,3-Di-O-acetyl-4-S-benzoyl-4-thio-β-L-ribopyranoside (XIIIb).—A solution of 0.25 g. (0.87 m*M*) of methyl 4-S-benzoyl-4-thio-β-L-ribopyranoside (XIIb) and 10 mg. of *p*-toluenesulfonic acid in 10 ml. of acetic anhydride was stored at room temperature for 18 hr. The reaction mixture was decomposed by adding it dropwise with stirring to 200 ml. of saturated aqueous sodium bicarbonate. The mixture was extracted with two 50-ml. portions of chloroform; the chloroform extracts were washed with water, then dried and evaporated to dryness *in vacuo* to give a quantitative yield of product as a colorless oil, $\lambda_{max(\mu)}^{film}$ 5.70 (acetate C=O), 5.98 (thiolbenzoyl C=O), 14.5 (thiolbenzoyl phenyl), $[\alpha]^{25}D$ +130° (1% in chloroform).

Anal. Calcd. for $C_{17}H_{20}O_7S$: C, 55.4; H, 5.44; S, 8.70. Found: C, 55.5; H, 5.40; S, 8.80.

1,2,3,5-Tetra-O-acetyl-4-thio-D-ribofuranose (XIVa).—To an ice-cold solution of 380 ml. of acetic acid, 380 ml. of acetic anhydride, and 23 ml. of concentrated sulfuric acid was added 16.4 g. of methyl 4-S-benzoyl-4-thio-β-D-ribopyranoside. The reaction mixture was stored at 0° for 48 hr., then was decomposed by the addition of 85 g. of anhydrous sodium acetate. The resulting mixture was evaporated to dryness *in vacuo* at a temperature below 30°. The residue was partitioned between 500 ml. of water and 200 ml. of chloroform. The aqueous phase was extracted with three 100-ml. portions of chloroform. The combined chloroform layers were evaporated to dryness *in vacuo*, then several portions of methanol were added and removed *in vacuo* to eliminate the last traces of acetic anhydride. The residue was dissolved in 200 ml. of chloroform and washed with three 100-ml. portions of saturated aqueous sodium bicarbonate. The chloroform layer was dried, then evaporated to dryness *in vacuo* to give 18.7 g. (97%) of product as a pale yellow sirup, $[\alpha]^{25}D$ −59° (1% in chloroform), $\lambda_{max(\mu)}^{film}$ 5.70 (acetate C=O) 8.20 (acetate C–O–C); there was no S-benzoate carbonyl at 6.0 μ or phenyl band at 14.5 μ.

Anal. Calcd. for $C_{13}H_{18}O_8S$: C, 46.7; H, 5.39; S, 9.59. Found: C, 46.8; H, 5.44; S, 9.75.

Crystallization from methanol gave a 31% yield of crystalline β-tetraacetate, m.p. 64.5–66°, $[\alpha]^{22}D$ −102° (1% in chloroform).

Anal. Found: C, 46.8; H, 5.64; S, 9.66.

1,2,3,5-Tetra-O-acetyl-4-thio-L-ribofuranose (XIVb) was prepared by the procedure described for the preparation of the D-isomer starting from methyl 4-S-benzoyl-4-thio-β-L-ribopyranoside (XIIb) to yield a yellow sirup, $[\alpha]^{25}D$ +56° (1.3% in chloroform).

Anal. Found: C, 46.5; H, 5.22; S, 9.81.

9-(4′-Thio-β-D-ribofuranosyl)adenine (Thioadenosine) (XXVIIIa).—A solution of 5.0 g. (14.7 m*M*) of 1,2,3,5-tetra-O-acetyl-4-thio-D-ribofuranose (XIVa) and 1 ml. of acetyl chloride in 150 ml. of dry ether was saturated with anhydrous hydrogen chloride at 0°. The solution was stored at 0° for 4 days, then evaporated to dryness *in vacuo* to give a quantitative yield of the sirupy chloro sugar XXVa. Condensation of the chloro sugar XXVa with chloromercuri-6-benzamidopurine in the way described for the preparation of XXVIIIb (see below) gave the blocked nucleoside XXVIa which was deacylated directly by heating for 1 hr. in 250 ml. of 0.1% methanolic sodium methoxide. The dark red solution was neutralized with acetic acid then evaporated to dryness *in vacuo*. The pink solid was triturated several times with ether, then with 25 ml. of cold water. The resulting solid was recrystallized twice from methanol to give 1.1 g. (26%) of product (XXVIIIa), m.p. 248–249°, $[\alpha]^{25}D$ −42° (0.5% in 50% aqueous pyridine); $\lambda_{max(m\mu)}^{pH\ 1}$ 259 (ε 14,290); $\lambda^{pH\ 7,13}$ 261 (ε 14,870). The product was homogeneous on paper chromatography and had $R_{Adenine}$ values of 0.90, 0.63, and 1.25 in solvents A, B, and C, respectively.

Anal. Calcd. for $C_{10}H_{13}N_5O_3S \cdot 0.5H_2O$: C, 41.1; H, 4.81; N, 24.0; S, 11.0. Found: C, 41.2; H, 4.80; N, 23.9; S, 11.0.

(2′,3′-O-Isopropylidene-4′-thio-β-D-ribofuranosyl)adenine (XXXIIIa).—To a solution of 1 g. of *p*-toluenesulfonic acid in 15 ml. of acetone was added 100 mg. of 4′-thioadenosine (XXVIIIa). After stirring for a few minutes at room temperature, solution was complete. The reaction was stirred for a total of 40 min., then 1 g. of solid sodium bicarbonate was added and the mixture was stirred an additional 2 hr. The mixture was heated for 5 min., then filtered. The filter cake was washed with acetone, then triturated with a small amount of water. The water-insoluble material was filtered to give 45 mg. (40%) of product, m.p. 282–284° dec. Purification was effected by dissolving the material in dimethyl sulfoxide and precipitating in water to give the analytical sample, m.p. 288–289° dec.

Anal. Calcd. for $C_{13}H_{17}N_5O_3S$: C, 48.3; H, 5.27; N, 21.7; S, 9.91. Found: C, 48.3; H, 5.30; N, 21.5; S, 10.0.

A larger run using 250 mg. of nucleoside gave a 78% yield of acetonate XXXIIIa, m.p. 288–290° dec.

4-Thio-L-ribofuranose (IVb) and the Disulfide XVIIIb.—A solution of 480 mg. of 1,2,3,5-tetra-O-acetyl-4-thio-L-ribofuranose (XIVb) in 10 ml. of methanol which contained 10 mg. of sodium methoxide was stirred at room temperature for 3 hr. The reaction was neutralized with Amberlite IRC-50 (H), then filtered through a Celite pad. The filtrate was evaporated to dryness *in vacuo* to give 0.2 g. (80%) of a colorless oil that gave a positive nitroprusside test for a thiol and which was homogeneous on thin-layer chromatography using solvent D. The absence of significant absorption at 3.90 and 5.75 μ in the infrared spectrum suggested that the material existed primarily as a thiofuranose. This sirup reacted rapidly with aqueous iodine solution at room temperature to give about the theoretical uptake for one thiol group, after which there was a relatively slow additional iodine consumption.

Anal. Calcd. for $C_5H_{10}O_4S$: C, 36.1; H, 6.03; S, 19.3. Found: C, 36.2; H, 6.30; S, 19.1.

In a second deacylation, 830 mg. of the tetraacetate XIVb in 15 ml. of methanol containing 10 mg. of sodium methoxide was left at room temperature for 60 hr. Neutralization with Amberlite IRC-50 (H) and evaporation of the filtrate to dryness gave 450 mg. of a white solid which gave a positive nitroprusside test but which had two components according to thin layer chromatography in solvent D. Trituration of the crude solid with methanol, then diethyl ether, gave 55 mg. of white solid, m.p. 179–183° dec., which gave a negative nitroprusside test and was homogeneous on thin-layer chromatography in solvent D.

Anal. Calcd. for $C_{10}H_{18}O_8S_2$: C, 36.3; H, 5.45; S, 19.4 Found: C, 36.4; H, 6.05; S, 19.10.

Methyl 2,3,5-Tri-O-(*p*-nitrobenzoyl)-4-thio-L-ribofuranoside (XVIIb).—A solution of 550 mg. of 4-thio-L-ribose (IVb) in 15 ml. of 0.5% methanolic hydrogen chloride was heated at reflux for 4 hr. The solution was cooled, then neutralized with IR-45 (OH) resin, and decolorized with Norit. The methanol solution was filtered through Celite and evaporated to dryness *in vacuo* to give crude methyl 4-thio-L-ribofuranoside (XVIb) as a colorless oil, which had no evidence for thiol absorption in the infrared and which gave a negative nitroprusside test and reducing sugar test.

A solution of 400 mg. of the glycoside XVIb in 20 ml. of dry pyridine was treated with 3.5 g. (18.9 m*M*) of *p*-nitrobenzoyl chloride for 4 days at room temperature. The reaction mixture was added dropwise with stirring to 200 ml. of saturated aqueous sodium bicarbonate and stirred until crystallization was complete. The crystals which were collected weighed 1.37 g. (99%), m.p. 105–108°. The analytical sample was prepared by recrystallization from methanol–chloroform (2:1) to give white needles with a double melting point, m.p. 108–110°, then 195.0–195.5°, $[\alpha]^{25}D$

$-89°$ (1% in chloroform); $\lambda_{max(\mu)}^{Nujol}$ 5.78 (C=O), 6.52, 7.40 (NO_2), 7.85 (benzoate C–O–C), 13.92 (C_6H_5).

Anal. Calcd. for $C_{27}H_{21}N_3O_{14}S \cdot H_2O$: C, 50.2; H, 3.56; N, 6.50; S, 4.95. Found: C, 50.2; H, 3.38; N, 6.04; S, 4.95.

1,2,3,5-Tetra-O-(*p*-nitrobenzoyl)-4-thio-L-ribofuranose (XVb).—A solution of 1.0 g. (6.0 m*M*) of 4-thio-L-ribofuranose (IVb) in 50 ml. of dry pyridine was treated with 10 g. (54 m*M*) of *p*-nitrobenzoyl chloride at room temperature for 18 hr. The reaction mixture was poured slowly with vigorous stirring into 200 ml. of saturated aqueous sodium bicarbonate. The aqueous suspension was extracted with three 50-ml. portions of chloroform. The combined chloroform layers were washed with water, then dried and evaporated to dryness *in vacuo* to leave a brown gummy solid.

The residue was triturated with 50 ml. of hot ethanol–chloroform (1:1) until all the gum had dissolved. The remaining white solid was filtered to yield 1.3 g. (28%) of white crystals, m.p. 195–215°. Recrystallization from chloroform gave the analytical sample, m.p. 218–220°, $[\alpha]^{25}D$ +30° (1.36% in chloroform), $\lambda_{max(\mu)}^{Nujol}$ 5.78 (ester C=O). The absence of absorption at 6.0 μ indicated there was no S-(*p*-nitrobenzoate).

Anal. Calcd. for $C_{33}H_{22}N_4O_{16}S$: C, 52.0; H, 2.89; N, 7.35; S, 4.20. Found: C, 52.0; H, 2.95; N, 7.28; S, 4.16.

A second crop of the tetra-*p*-nitrobenzoate was obtained by flooding the ethanol–chloroform filtrate with Skellysolve B to obtain 1.5 g. (33%) of white precipitate, m.p. *ca.* 130°, which had an infrared spectrum compatible with the expected structure. This material had $[\alpha]^{25}D$ −33° (1% in 1,2-dichloroethane) which suggests that the crystalline high-melting tetranitrobenzoate has the β-configuration.

2,3,5-Tri-O-(*p*-nitrobenzoyl)-4-thio-L-ribofuranosyl Bromide (XXIVb).—To a solution of 0.5 g. of 1,2,3,5-tetra-O-(*p*-nitrobenzoyl)-4-thio-L-ribofuranose (XVb) in 1,2-dichloroethane was added 10 ml. of 30% anhydrous hydrogen bromide in glacial acetic acid. The reaction was stirred at room temperature for 18 hr. and the resulting solid was removed by filtration and washed with 3 ml. of cold acetic acid, then with five 5-ml. portions of dry diethyl ether. The resulting solid weighed 0.3 g. (68%) and had m.p. 175.0–176.5°, $[\alpha]^{24}D$ −38° (1% in 1,2-dichloroethane); $\lambda_{max(\mu)}^{Nujol}$ 5.75, 5.78 (ester C=O), 6.60, 7.40 (NO_2), 7.80, 7.90 (benzoate C–O–C), 8.92, 9.05, 9.15, 9.45 (sugar C–O–C).

Anal. Calcd. for $C_{26}H_{18}BrN_3O_{12}S$: C, 46.2; H, 2.68; Br, 11.8; N, 6.21; S, 4.73. Found: C, 46.0; H, 3.01; Br, 11.5; N, 6.20; S, 4.90.

Treatment of the high melting tetranitrobenzoate XVb under the same conditions gave identical results.

4′-Thio-L-adenosine (XXVIIIb). A. From Acetolysis of XIIb.—A mixture of 4.2 g. (6.2 m*M*) of 2,3,5-tri-O-(*p*-nitrobenzoyl)-4-thio-L-ribofuranosyl bromide (XXIVb) and 9.0 g. (10 m*M*) of chloromercuri-6-benzamidopurine (containing 4.5 g. of Celite) was heated at reflux in 375 ml. of dry xylene for 6 hr. The hot reaction mixture was filtered and the filtrate was diluted with 1 l. of Skellysolve B. The precipitated blocked nucleoside was dissolved in 250 ml. of dichloromethane. The dichloromethane solution was washed with two 100-ml. portions of 30% aqueous potassium iodide, then two 100-ml. portions of water. The organic layer was dried, then evaporated to dryness *in vacuo* to yield the crude blocked nucleoside as a yellow foam.

A solution of the crude blocked nucleoside in 250 ml. of methanol, which had been saturated with ammonia at 0°, was stored at room temperature for 24 hr., then evaporated to dryness *in vacuo*. The residue was crystallized from methanol to give 295 mg. (17%) of white crystalline solid, m.p. 248.0–249.0°.

The analytical sample from a previous reaction was recrystallized from methanol and had m.p. 248.5–249.5°, $[\alpha]^{23}D$ +41° (0.518% in 50% aqueous pyridine), $\lambda_{max(m\mu)}^{pH\ 1}$ 259 (ϵ 14,540), $\lambda_{max}^{pH\ 7.13}$ 261 (ϵ 14,890). The paper chromatography behavior was identical with that of D-thioadenosine.

Anal. Calcd. for $C_{10}H_{13}N_5O_3S$: C, 42.4; H, 4.62; N, 24.7; S, 11.3. Found: C, 42.6; H, 4.79; N, 24.6; S, 11.5.

B. From Acetolysis of XXX.—Benzoylation of 1.0 g. of methyl 4-S-benzoyl-4-thio-β-L-ribopyranoside (XIIb) in 10 ml. of pyridine with 1.5 ml. of benzoyl chloride gave a 92% yield of tribenzoate XXX as a sirup which had $\lambda_{max(\mu)}^{film}$ 5.75 (benzoate C=O), 5.98 (thiobenzoate C=O), 7.85–8.0 (benzoate C–O–C), 14.05 ($OOCC_6H_5$), 14.52 ($-S(O=)CC_6H_5$); there was no hydroxyl absorption at 2.9 μ. The n.m.r. spectrum showed a ratio of aromatic protons/methoxyl protons of 15:3.

A solution of 1.2 g. of the tribenzoate XXX in 27.5 ml. of acetic anhydride, 27.5 ml. of acetic acid, and 1.6 ml. of concentrated sulfuric acid was kept at 0° for 3 days, then decomposed and worked up in the manner described for the preparation of D-thioribose tetraacetate XIVa to give 800 mg. (73%) of tan colored sirup; $\lambda_{max(\mu)}^{film}$ 5.80 (benzoate C=O), 7.85 (benzoate C–O–C), 8.10, 8.20 (acetate C–O–C). There was no evidence for S-benzoate absorptions at 6.0 and 14.5 μ. The n.m.r. spectrum showed a ratio of aromatic protons to acetate protons of 10:6 as would be expected for the ribose derivative XXXI. From the infrared and n.m.r. spectral data it can be seen there was no loss of O-benzoate and complete loss of S-benzoate during the acetolysis of XXX.

Treatment of the acetolysis product XXXI with hydrogen chloride in ether followed by the usual nucleoside condensation with chloromercuri-6-benzamidopurine gave, after deacylation, L-thioadenosine, m.p. 246–248°, which did not depress in melting point on admixture with product from procedure A and which was identical in all respects with the product from procedure A.

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