Dinh, J. Org. Chem., 40, 2825 (1975).
(3) T. Huynh-Dinh, J. Igolen, E. Bisagni, J.-P. Marquet, and A. Civier, results to be published.
(4) J. D. Bower, J. Chem. Soc., 4510 (1957).
(5) K. T. Potts and H. R. Burton, J. Org. Chem., 31, 251 (1966).
(6) J. D. Bower and F. P. Doyle, J. Chem. Soc., 727 (1957).
(7) S. Naqui and V. R. Srinwasan, Indian J. Chem., 3, 162 (1965).
(8) M. S. Gibson, Tetrahedron, 19, 1587 (1963).
(9) K. T. Potts and S. R. Surapaneni, J. Heterocycl. Chem., 7, 1019 (1970).
(10) T. Okamoto, M. Hibobe, and E. Yabe, Chem. Pharm. Bull., 14, 523 (1966).
(11) T. J. Pugmire, M. J. Robins, D. M. Grant, and R. K. Robins, J. Am. Chem. Soc., 93, 1887 (1971).
(12) T. Novinson, T. Okabe, R. K. Robins, and P. Dea, J. Heterocycl. Chem., 12, 1187 (1975).
(13) (a) L. B. Townsend in "Synthetic Procedures in Nucleic Acid Chemistry", Vol. 2, W. W. Zorbach and R. S. Tipson, Ed., Wiley-Interscience, New York, N.Y., 1973, p 267; (b) G. Trummlitz, D. B. Repke, and J. G. Moffatt, J. Org. Chem., 40, 3352 (1975).
(14) J. L. Imbach, Ann. N.Y. Acad. Sci., 225, 177 (1975).
(15) A. H. Cook, A. C. Davis, I. Heilbron, and G. H. Thomas, J. Chem. Soc., 1071 (1949).
(16) H. Bader, J. D. Downer, and P. Driver, J. Chem. Soc., 2775 (1950).
(17) After completion of this report, a communication of Fox et al. [J. Heterocycl. Chem., 13, 175 (1976)] described the preparation of 8-(D-ribofuranosyl)-pyrazolo[1,5-a]-1,3,5-triazine.

6-Oxa Analogues of Pyrimidines and Pyrimidine Nucleosides. Synthesis of 5-Amino-6*H*-1,2,4-oxadiazin-3(2*H*)-one, 2-β-D-Ribofuranosyl-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione, and Related Derivatives

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Treatment of 6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**1**, 6-oxadihydrouracil) with phosphorus pentasulfide in dioxane gave 6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (**2**, 4-thio-6-oxadihydrouracil). Amination of **2** with ammonia in dioxane gave 5-amino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**3**, 6-oxadihydrocytosine). Treatment of **2** with dimethylamine in dioxane afforded 5-dimethylamino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**5**). The stannic chloride catalyzed condensation of 3,5-bis(trimethylsilyloxy)-6*H*-1,2,4-oxadiazine (**6**) and 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-β-D-ribofuranose (**7a**) or 1,2,3,5-tetra-*O*-acetyl-β-D-ribofuranose (**7b**) gave the corresponding blocked 6-oxadihydrouridines 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8a**) and 2-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8b**). 2-β-D-Ribofuranosyl-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8c**, 6-oxadihydrouridine) was obtained by the removal of the acetyl blocking groups of **8b** with methanolic hydrogen chloride. Thiation of **8a** with phosphorus pentasulfide in dioxane afforded 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (**12**), which upon treatment with dimethylamine in dioxane gave 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-5-dimethylamino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**13**). The stannic chloride catalyzed condensation of 3-trimethylsilyloxy-5-dimethylamino-6*H*-1,2,4-oxadiazine (**14**) with **7a** also afforded **13**. The ^{13}C NMR spectra of several of the above 6*H*-1,2,4-oxadiazin-3(2*H*)-ones are reported and have been utilized to support structural assignments.

6*H*-1,2,4-Oxadiazine-3,5(2*H*,4*H*)-dione (**1**) and 6-methyl-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione, 6-oxa analogues of uracil and thymine, respectively, have previously been synthesized.[1] These analogues are actually isosteres of 5,6-dihydrouracil and 5,6-dihydrothymine in which the 6-methylene group has been replaced by an oxygen such that these compounds can be considered as 6-oxadihydrouracil (**1**) and 6-oxadihydrothymine. It has been shown, however, that 6-oxadihydrouracil (**1**) is an apparent competitive antagonist of uracil, and not of dihydrouracil, in bacterial systems.[2] In an effort to further investigate the chemical and biochemical properties of the 6*H*-1,2,4-oxadiazin-3(2*H*)-one ring system, we have synthesized the 6-oxa analogues of 4-thiouracil, cytosine, and *N*,*N*-dimethylcytosine as well as the 6-oxa analogue of uridine, the first 6*H*-1,2,4-oxadiazine nucleoside.

Reaction of 6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**1**) with phosphorus pentasulfide in refluxing, anhydrous dioxane afforded 6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (**2**, 4-thio-6-oxadihydrouracil) in 55% yield. Elemental analysis established that **2** was a monothio derivative of **1**. The shift in the uv maximum from 220 nm (ε 1250) to 272 nm (ε 15 900) upon thiation was similar to that found upon thiation of 5,6-dihydrouracils.[3] That the 6*H*-1,2,4-oxadiazine ring had remained intact was shown by the almost quantitative reconversion of **2** to **1** by boiling water. Thiation of **1** was expected to give the 5-thio derivative in analogy to the thiation of 5,6-dihydrouracils.[3] Unequivocal assignment of the structure of 6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (**2**) is based on subsequent transformation of **2** to 5-amino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**3**) as described below.

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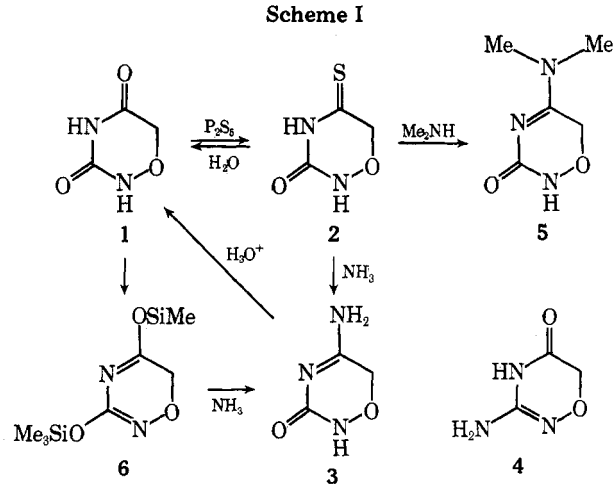
Scheme I



Reaction of 6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (**2**) with ammonia in dioxane at room temperature resulted in conversion to 5-amino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**3**, 6-oxadihydrocytosine) in 90% yield. The highly reactive nature of the thio group of **2** is analogous to that found for 1-alkyl-

4-thio-5,6-dihydrouracils.[4] The shift in the uv maximum from 220 nm (ε 1250) to 228 nm (ε 13 200) upon amination and the much lower frequency (1620 cm^{-1}) of the C-3 carbonyl absorption in the ir spectrum of **3** as compared to that of **1** (1745 and 1710^{-1} cm) is indicative of the conjugation of the C-3 carbonyl with the 4,5 double bond of **3**. A similar shift is seen upon comparison of 5,6-dihydrouracil and 5,6-dihydrocytosine.[4]

Deamination of 5-amino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**3**) to 6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**1**) by treatment with dilute acid at room temperature confirmed that the 6*H*-1,2,4-oxadiazine ring had remained intact during amination. The 5-amino rather than the 3-amino structure was assigned to **3** since this product was shown to be different from an authentic sample of the known 3-amino-6*H*-1,2,4-oxadiazin-5(4*H*)-one (**4**)[1] by comparison of ir, uv, and ^{1}H NMR spectra as well as melting point and TLC. Since 5-amino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**3**) was derived from 6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (**2**), the assignment of **2** as the 5-thio derivative is thereby firmly established.

Reaction of 6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (**2**) with dimethylamine in dioxane at room temperature resulted in conversion to 5-dimethylamino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**5**) in 87% yield.

It was also found that 6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**1**) could be converted in 20% yield to 5-amino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**3**) by conversion in situ to 3,5-bis(trimethylsilyloxy)-6*H*-1,2,4-oxadiazine (**6**) and subsequent reaction with ammonia.[6,7]

The ^{13}C NMR spectra of several 6*H*-1,2,4-oxadiazin-3(2*H*)-ones have been obtained and the chemical shifts are summarized in Table II. The relative ordering of the C-3 and C-5 carbons was assigned by comparing the chemical shifts of 6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**1**) with those of the anion formed by LiOH in Me_2SO-d_6. Since the negative charge is expected to be localized at the oxygen atom of the C-3 carbonyl, the C-3 resonances of **1** and the corresponding anion should differ more than the C-5 resonances. The observed shift difference of 11.9 ppm for the C-3 carbons and the 2.8-ppm difference for the C-5 carbons strongly supports the assignment of C-3 and C-5 as indicated in Table II. Additional confirmation for the assignment of the C-3 and C-5 carbons of **1** was obtained by comparison with the ^{13}C NMR spectra of uridine,[8] where the resonance for the C-4 carbon occurs downfield from the C-2 carbon.

The significant downfield shift observed in the C-5 resonance (33.6 ppm) of 6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (**2**) confirms that thiation had occurred at C-5. Similar downfield shifts upon substitution of sulfur for oxygen have been observed for the thiopyrimidine nucleosides.[8] In the ^{13}C NMR spectrum of 5-amino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**3**), the C-5 resonance occurs downfield from the C-3 resonance, analogous to the ^{13}C NMR spectrum of cytidine[8] where the C-4 resonance occurs downfield from the C-2 resonance.

After 5-amino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**3**) was heated in deuterium oxide for 24 h at 55 °C (necessary for complete dissolution), the ^{1}H NMR spectrum indicated incorporation of deuterium at the C-6 position. Some hydrolysis to 6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**1**) had occurred as indicated by TLC and the appearance in the ^{1}H NMR spectrum of a signal for the C-6 protons of **1**. Under the same conditions the C-6 protons of **1** did not exchange. In pH 7.5 Tris buffer at room temperature, **3** slowly under went hydrolysis ($t_{1/2}$ = 5 days) to **1**. The facile hydrolysis of 5-amino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**3**) to 6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**1**) and the exchange of the C-6 protons of **3** with deuterium oxide are analogous to the results found for 5,6-dihydrocytosine.[4] The similarity of the amino group of **3** with that of 5,6-dihydrocytosine is indicated by the similarity of the pK_a values of these two compounds. The pK_a of **3** is 6.1 while that of 5,6-dihydrocytosine is 6.3.[4]

Table I. Properties of 6*H*-1,2,4-Oxadiazin-3(2*H*)-ones

Compd	pK_a	Uv (EtOH), λ_{max}, nm ($\epsilon \times 10^{-3}$)	Ir, cm^{-1} [a]
1	7.6	220 (1.25)	3170; 3070; 1745; 1710
2	7.1	274 (15.9)	3190; 1720
3	6.1	228 (13.2)	3250; 1620
4		234 (7.78)	3200; 3120; 1660; 1620
5		245 (16.4)	3110; 1645
8a		230 (44.1)	3240; 1730
8b		217 (1.65)	3210; 3100; 1760; 1725
8c	8.7	219 (1.80)	3360; 1710
10	9.1	225 (1.64)	3170; 3070; 1725
11	7.8	*b*	3250; 1735; 1680
12		276 (22.0)	3230; 3180; 1745; 1715
		229 (40.9)	
13		250 (21.9)	1735; 1675
		231 (46.3)	

[a] KBr. [b] End absorption only.

We next investigated the synthesis of 2-β-D-ribofuranosyl-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8c**, 6-oxadihydrouridine). Reaction of 6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**1**) in refluxing dioxane with hexamethyldisilazane (HMDS) in the presence of ammonium sulfate afforded 3,5-bis(trimethylsilyloxy)-6*H*-1,2,4-oxadiazine (**6**), which without further purification was condensed in 1,2-dichloroethane with 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-β-D-ribofuranose (**7a**) and 1 equiv of stannic chloride[9] to afford a 68% yield of 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8a**), the only nucleoside product detected by TLC. The ^{1}H NMR signal for the anomeric proton of **8a** appeared as a singlet, and coupling constants of less than 1.0 Hz establish the β configuration[10] for ribonucleosides. It was also possible to prepare **8a** by the reaction of 2,3,5-tri-*O*-benzoyl-D-ribofuranosyl bromide (**9**) in acetonitrile with **6**, with the sodium salt of **1**, or by the reaction of **9** with **1** in nitromethane in the presence of mercuric cyanide.[11] In these cases, however, the yield of **8a** was considerably reduced.

Treatment of 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8a**) with sodium methoxide in methanol gave a different product than that obtained by the treatment with methanolic ammonia. In neither case was the desired 2-β-D-ribofuranosyl-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8c**) obtained. While the benzoyl groups were removed in each case, ring opening probably had occurred as noted by the lack of uv absorbance of the resulting products, which were not further investigated.

As the 6*H*-1,2,4-oxadiazine ring of 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8a**) was unstable to the above basic deblocking conditions, and since the acetyl blocking groups of a ribonucleoside can be removed by the use of acidic conditions,[12] 2-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5-(2*H*,4*H*)-dione (**8b**) was synthesized from 1,2,3,5-tetra-*O*-acetyl-β-D-ribofuranose (**7b**) and **6** in 86% yield using the same conditions as for the synthesis of **8a**. Treatment of 2-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5-(2*H*,4*H*)-dione (**8b**) with anhydrous methanolic hydrogen chloride afforded 2-β-D-ribofuranosyl-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8c**, 6-oxadihydrouridine) in 79% yield. Support for the assignment of the site of ribosylation as N-2 rather than N-4 is based on a comparison of the pK_a of 6-oxadihydrouridine (**8c**) with that of 2-methyl-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**10**)[5] and 4-methyl-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**11**)[5] (Table I). The value of 8.7

Table II. ^{13}C Chemical Shifts of Some 6*H*-1,2,4-Oxadiazin-3(2*H*)-ones

Compd	Chemical shift, ppm[a]			
	C-3	C-5	C-6	NCH_3
1	155.7	169.4	69.3	
Anion of 1	167.6	172.2	67.6	
2	151.4	203.0	75.9	
3	162.4	174.8	64.2	
8b	153.1	168.9	70.4	
10	155.2	169.2	69.7	35.0
11	155.8	168.2	69.8	25.5

[a] Chemical shifts are measured from Me_2SO-d_6, and are converted to Me_4Si scale using the relationship δ Me_4Si = δ Me_2SO-d_6 + 39.5 ppm.

found for the pK_a of **8c** is much closer to the value of 9.1 found for the pK_a of the 2-methyl derivative **10** than the value of 7.8 found for the pK_a of the 4-methyl derivative **11**. Proof for the assignment of the site of ribosylation was possible from subsequent transformations.

Comparison of the ^{13}C NMR spectra of 2-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8b**), **10**, and **11** (Table II) indicates that the site of ribosylation cannot be assigned on the basis of these data. The substitution of a methyl group or a β-D-ribofuranosyl moiety for the proton at the N-2 or N-4 position of 6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**1**) has little effect on the carbon-13 chemical shift values of the C-3 or C-5 carbons. This suggests that the structures of these compounds are essentially unchanged upon substitution of the NH proton and that the keto form predominates in both cases.

In aromatic heterocycles, N-substitution has been observed to produce a significant upfield shift in the ^{13}C NMR signal of the carbon α to the substituted nitrogen and a downfield shift in the signal of the carbon β to that nitrogen[13] when the neutral species is compared with the corresponding anion. Owing to the lack of aromaticity in these compounds, the negative charge of the anion is not delocalized around the ring and the corresponding substitution shifts are therefore not observed.

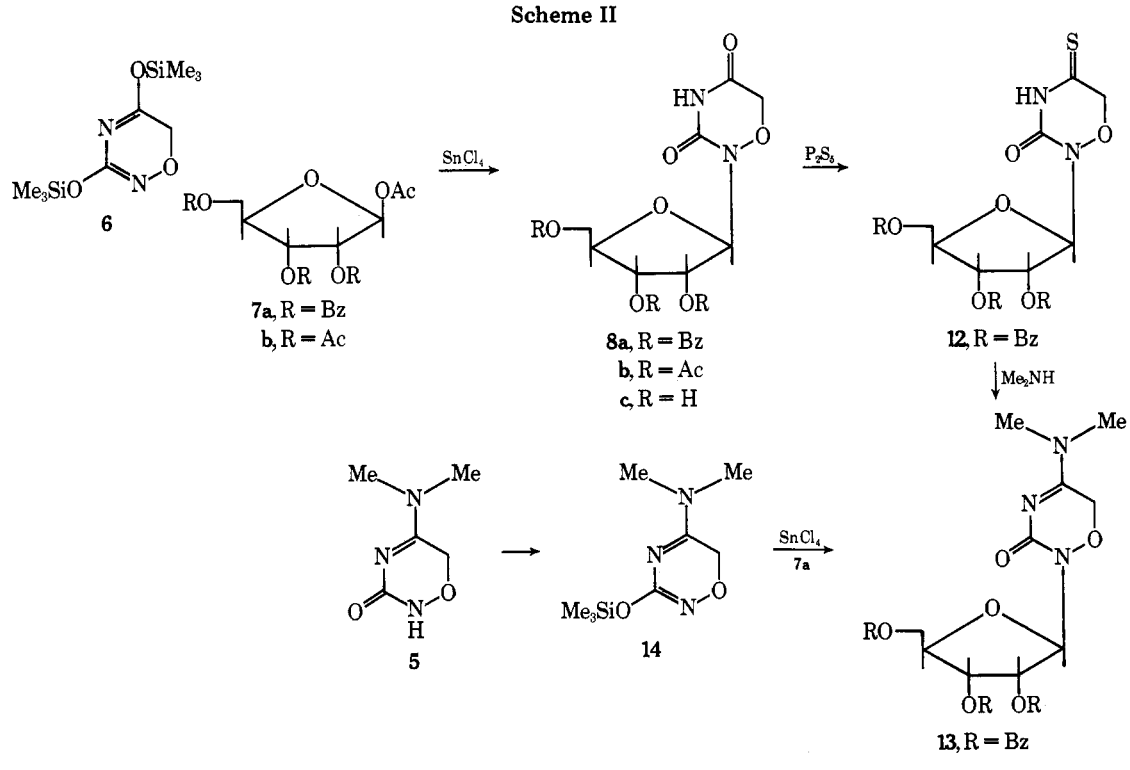
It is interesting to note that the ^{13}C chemical shift of the *N*-methyl carbon of 2-methyl-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**10**) occurs considerably more downfield than that of 4-methyl-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**11**). In order to see if this β effect of a carbonyl group on an *N*-methyl carbon is of a general nature, we investigated the ^{13}C NMR spectrum of 1,3-dimethyluracil, which had been previously reported without assignment of the *N*-methyl carbons.[14] By examining the proton coupled ^{13}C NMR spectrum of 1,3-dimethyluracil in Me_2SO-d_6 it was possible to assign the resonance at 36.8 ppm to the N-1 methyl carbon and the resonance at 27.5 ppm to the N-3 methyl carbon based on the small vicinal coupling (5 Hz) of the C-6 proton to the N-1 methyl carbon. Therefore, the ^{13}C chemical shift of the N-3 methyl carbon of 1,3-dimethyluracil, which is adjacent to two carbonyl groups, also occurs at a considerably higher field than that of the N-1 methyl carbon, which is adjacent to only one carbonyl group.

In an effort to provide further proof for the site of ribosylation of 2β-D-ribofuranosyl-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8c**), we undertook the synthesis of 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (**12**) and 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-5-dimethylamino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**13**).

Thiation of 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8a**) with phosphorus pentasulfide in anhydrous, refluxing dioxane afforded 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (**12**) in 45% yield. Elemental analysis established that **12** was a monothio derivative of **8a** and, as seen upon thiation of 6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**1**), the uv maxima of **12**, 229 nm (ε 40 900) and 276 (22 000), had shifted as compared to the uv maximum of 230 nm (ε 44 100) for **8a**.

The ^1H NMR signal for the anomeric proton of 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazin-3-(2*H*)-one-5(4*H*)-thione (**12**) appeared at 6.18 ppm while that

Scheme II



of 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8a**) appeared at 6.12 ppm. The anisotropic effect of a thione group adjacent to the site of glycosylation causes a large shift of the ^{1}H NMR signal for the anomeric proton to lower field.[10,15,16] If **12** were the 3-thio rather than the 5-thio derivative, the ^{1}H NMR signal of the anomeric proton of **12** would be expected to appear at a much lower field than that of **8a**. The same would be true if **12** were the N-4 isomer, as then the site of glycosylation would be adjacent to both a carbonyl and a thione group. This indicates that **12** is the 5-thio rather than the 3-thio derivative, and, also, that both **8a** and **12** are the N-2 rather than the N-4 ribonucleosides.

Treatment of 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (**12**) with sodium methoxide in methanol gave a complex reaction mixture, which was not further investigated. Presumably the 6*H*-1,2,4-oxadiazine ring of **12** is unstable to basic deblocking conditions, as found for **8a**. Treatment of certain thio analogues of 5,6-dihydrouracil and their methyl derivatives with sodium methoxide in methanol also resulted in ring opening.[17]

Reaction of 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (**12**) with dimethylamine in dioxane at room temperature afforded 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-5-dimethylamino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**13**) in 53% yield. It was also possible to prepare **13**, in 61% yield, by condensation in 1,2-dichloroethane of 3-trimethylsilyloxy-5-dimethylamino-6*H*-1,2,4-oxadiazine (**14**) with 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-β-D-ribofuranose (**7a**) and 1 equiv of stannic chloride.[9] Silyl derivative **14** was prepared by treatment of 5-dimethylamino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**5**) in refluxing dioxane with HMDS in the presence of ammonium sulfate, and, after removal of solvents, was used without further purification.

The synthesis of 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-5-dimethylamino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**13**) from 5-dimethylamino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (**5**) firmly establishes **13** as the N-2 ribonucleoside. As 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazin-3-(2*H*)-one-5(4*H*)-thione (**12**) was converted to **13** and 2-(2,3,5-tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (**8a**) was converted to **12**, the assigned structures for **8a** and **12** thus received further support.

Experimental Section

Melting points were taken on a Thomas-Hoover capillary melting point apparatus and are uncorrected. Specific rotations were measured in a 1-dm tube with a Perkin-Elmer Model 141 automatic digital readout polarimeter. Nuclear magnetic resonance (^{1}H NMR) spectra were recorded at 60 MHz on a Hitachi Perkin-Elmer R-20A spectrometer in Me_2SO-d_6 using DSS as an internal standard. The ^{13}C NMR spectra were obtained on a Bruker HX-90 NMR spectrometer operating at 22.62 MHz in the Fourier transform mode at a probe temperature of 35 °C. A Fabri-Tek 1074 signal averager with 4096 word memory was used for data accumulation and a PDP-8/e computer for data processing. Solutions (1.0 M) were prepared in Me_2SO-d_6 and were studied in 10-mm tubes. Ultraviolet spectra (uv, $\epsilon \times 10^{-3}$) were recorded on a Cary Model 15 spectrophotometer and infrared spectra (ir) on a Perkin-Elmer 257 spectrophotometer (KBr pellets). Elemental analyses were performed by Galbraith Laboratories, Inc., Knoxville, Tenn. The pK_a determinations were performed on a Radiometer automatic potentiometric titrator. Evaporations were carried out under reduced pressure below 40 °C. Detection of components on silica gel (ICN, Woelm F254) was by ultraviolet light and with anisaldehyde spray followed by heating.

6*H*-1,2,4-Oxadiazin-3(2*H*)-one-5(4*H*)-thione (2). A solution of 23.2 g (200 mmol) of **1** and 23.2 g (100 mmol) of purified P_2S_5 in 1000 ml of dry dioxane was refluxed for 2 h. After cooling, the reaction mixture was filtered and the filtrate concentrated in vacuo to about 300 ml. Silica gel (80 g) was added and the solvent removed in vacuo. The residue was applied to an 800-g silica gel dry column (2.75 in. nylon tubing) followed by 200 g of W200 alumina. The column was eluted with 2000 ml of 6:4 CH_2Cl_2–Et_2O and 5000 ml of 1:1 CH_2Cl_2–Et_2O, fractions of 500 ml being collected. Fractions 2–9 were combined and after removal of solvent in vacuo the crude product was recrystallized from CH_3CN to give 10.77 g. Recrystallization from CH_3CN gave the analytical sample: mp 153.5–154.5 °C; NMR (Me_2SO-d_6) δ 4.68 (s, 2, CH_2), 11.4 (br s, 1, NH), 13.6 (br s, 1, NH).

Anal. Calcd for $C_3H_4N_2O_2S$ (132.141): C, 27.27; H, 3.05; N, 21.20; S, 24.26. Found: C, 27.47; H, 3.06; N, 21.14; S, 24.39.

Workup of the mother liquor afforded 3.83 g of additional product for a total yield of 14.60 g (55.3%).

5-Amino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (3). A. Dry ammonia was bubbled into a solution of 1.33 g (10 mmol) of **2** in 50 ml of dry dioxane for 2.5 h. The yellow suspension was filtered and the filter cake washed with dry dioxane (2 × 10 ml). After washing well with $CHCl_3$ there remained 1.039 g of white solid (90.3%). Recrystallization from MeOH followed by two recrystallizations from EtOH gave the analytical sample: mp 151–152 °C; NMR (Me_2SO-d_6) δ 4.33 (s, 2, CH_2), 7.9 (broad s, 1, NH), 9.1 (broad s, 2, NH_2).

Anal. Calcd for $C_3H_5N_3O_2$ (115.092): C, 31.31; H, 4.38; N, 36.51. Found: C, 31.20; H, 4.18; N, 36.40.

B. Ammonia was bubbled into a solution of 10 ml of HMDS and 50 ml of dry dioxane for 30 min at room temperature. This solution was then transferred to a bomb containing 1.16 g (10 mmol) of **1** and 100 mg of ammonium sulfate. The bomb was heated on a steam bath for 15 h. After cooling, the contents of the bomb were removed and the bomb washed well with $CHCl_3$. The solvents were removed in vacuo and the residue dried at the vacuum pump for 30 min. The residue was then suspended in EtOAc and filtered, and the solid was washed with EtOAc to give 0.235 g (20.4%) of slightly impure **3a** as determined by comparison with the TLC, uv, and ir spectrum of the material prepared by method A above.

EtOH was added to the EtOAc filtrate and solvents were then removed in vacuo. EtOH was added to the residue and then removed in vacuo to afford a dark brown solid residue, which was shown by TLC (7:3 $CHCl_3$–MeOH) to only contain more **3** as well as unreacted **1**.

5-Dimethylamino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (5). Dimethylamine was bubbled into a solution of 0.375 g (2.8 mmol) of **2** in 11 ml of dry dioxane for 5 min. After stirring at room temperature for 50 min, the resulting precipitate was filtered and washed with a little dioxane and then washed well with ether to give 0.350 g (87.3%) of white solid. Recrystallization from EtOH gave the analytical sample: mp 176–177 °C; NMR (Me_2SO-d_6) δ 3.03 and 3.08 (s, 6, NMe_2), 4.60 (s, 2, CH_2), 9.61 (broad s, 1, NH).

Anal. Calcd for $C_5H_9N_3O_2$ (143.146): C, 41.95; H, 6.34; N, 29.36. Found: C, 42.24; H, 6.63; N, 29.60.

2-(2,3,5-Tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5-(2*H*,4*H*)-dione (8a). A solution of 2.55 g (22 mmol) of **1** (powdered and dried for 1 day in vacuo over P_2O_5 at 80 °C), 220 mg of ammonium sulfate, and 20 ml of HMDS in 100 ml of dry dioxane was refluxed for 18 h. After cooling, dioxane and HMDS were removed in vacuo and the residue was then dried at the vacuum pump for 1 h. The semisolid residue was taken up in 200 ml of dry 1,2-dichloroethane (dried by a Woelm W200 basic alumina column and then stored overnight over 4A molecular sieves) after which 10 g of 4A molecular sieves and 10.08 g (20 mmol) of 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-β-D-ribofuranose (powdered and dried for 1 day in vacuo over P_2O_5 at 80 °C) were added. The flask was flushed with dry nitrogen and stoppered with a rubber septum. After $SnCl_4$ (2.3 ml, 20 mmol) was added via a syringe, the reaction mixture was stirred at room temperature for 22 h. The reaction mixture was then poured into 100 ml of saturated aqueous $NaHCO_3$ solution. After stirring well, Celite was added followed by filtration through a Celite pad. After the filter cake was washed well with CH_2Cl_2, the organic phase was washed with 50 ml of saturated aqueous NaCl solution and then dried over Na_2SO_4. Removal of the solvent in vacuo gave 9.60 g of a yellowish foam. Recrystallization from 250 ml of EtOH gave 6.846 g of white needles. A second recrystallization from EtOH gave the analytical sample: mp 174–174.5 °C; $[\alpha]^{25}D$ −38.3° (*c* 1.0, $CHCl_3$); NMR (Me_2SO-d_6) δ 4.50 (s, 2, CH_2), 6.12 (s, 1, $H_{1'}$).

Anal. Calcd for $C_{29}H_{24}N_2O_{10}$ (560.515): C, 62.14; H, 4.32; N, 5.00. Found: C, 62.23; H, 4.52; N, 4.76.

The mother liquor was removed in vacuo and the residue chromatographed on 300 g of dry column silica gel, eluting with 9:1 $CHCl_3$–EtOAc to give 0.763 g more product for a combined yield of 7.609 g (67.8%).

2-(2,3,5-Tri-*O*-acetyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (8b). A solution of 6.38 g (55 mmol) of **1**

(powdered and dried in vacuo at 60 °C for 6 h), 500 mg of ammonium sulfate, and 50 ml of HMDS in 250 ml of dry dioxane was refluxed for 15 h. After cooling, dioxane and HMDS were removed in vacuo and the residue was dried at the vacuum pump for 1 h. The semisolid residue was taken up in 350 ml of dry 1,2-dichloroethane after which 35 g of 4A molecular sieves and 16.0 g (50 mmol) of 1,2,3,5-tetra-*O*-acetyl-β-D-ribofuranose (powdered and dried in vacuo at 80 °C for 15 h) were added. The flask was flushed with nitrogen and stoppered with a rubber septum. After $SnCl_4$ (5.8 ml, 50 mmol) was added via a syringe, the reaction mixture was stirred at room temperature for 24 h. The reaction mixture was then poured into 250 ml of saturated aqueous $NaHCO_3$. After stirring well, Celite was added followed by filtration through a Celite pad. After the filter cake was washed well with $CHCl_3$, the organic phase was washed with 200 ml of saturated aqueous NaCl solution and then dried over $MgSO_4$. Removal of the solvent in vacuo followed by drying of the residue in vacuo for 15 h and then a further drying in vacuo over P_2O_5 for 24 h gave 16.1 g (86.0%) of a white glass, which was shown by TLC (85:15 $CHCl_3$–Me_2CO) to contain only minor impurities. Chromatography of a portion of the above on silica gel, eluting with 99:1 $CHCl_3$–MeOH, afforded an analytical sample: mp 45 °C; $[\alpha]^{25}D$ −24.3° (*c* 1.2, $CHCl_3$); NMR (Me_2SO-d_6) δ 4.71 (s, 2, CH_2), 5.80 (d, J = 4 Hz, 1, $H_{1'}$).

Anal. Calcd for $C_{14}H_{18}N_2O_{10}\cdot H_2O$ (392.321); C, 42.86; H, 5.14; N, 7.14. Found: C, 42.81; H, 5.36; N, 6.95.

2β-D-Ribofuranosyl-6*H*-1,2,4-oxadiazine-3,5(2*H*,4*H*)-dione (8c). A solution of 1.51 g (3.8 mmol) of **8b** in 75 ml of anhydrous 0.1 M MeOH–HCl was refrigerated for 25 h in a stoppered flask. The reaction mixture was neutralized with IR-45(OH) resin, and the resin was filtered and washed with MeOH. Removal of the MeOH in vacuo followed by drying the residue overnight in vacuo afforded 0.75 g (79.6%) of slightly impure **13** as a syrup. A homogeneous sample of **13** was obtained by preparative TLC on silica gel (7:3 $CHCl_3$–MeOH). The product was extracted from the silica gel with MeOH. Removal of the MeOH in vacuo gave a slightly yellowish syrup which was taken up in water, treated with charcoal, filtered through Celite, and then lyophilized to give a white solid: mp 180 °C dec; $[\alpha]^{25}D$ −14.2° (*c* 1.0, H_2O); NMR (Me_2SO-d_6) δ 4.63 (s, 2, CH_2), 5.52 (d, J = 5 Hz, 1, $H_{1'}$), 6.92 (br s, 1, NH).

Anal. Calcd for $C_8H_{12}N_2O_7\cdot H_2O$ (266.209): C, 36.09; H, 5.30; N, 10.52. Found: C, 36.60; H, 5.43; N, 10.43.

2-(2,3,5-Tri-*O*-benzoyl-β-D-ribofuranosyl)-6*H*-1,2,4-oxadiazin-3(2*H*)-one-5(4*H*)-thione (12). A solution of 1.122 g (2.0 mmol) of **8a** and 0.266 g (1.2 mmol) of P_2S_5 in 20 ml of dry dioxane was refluxed for 2 h. After cooling, the reaction mixture was filtered and the dioxane removed in vacuo to give 1.566 g of a yellow foam. Chromatography on silica gel (50 g), eluting with 98:2 CH_2Cl_2–Et_2O, gave 0.519 g (45.0%) of yellow solid. Two recrystallizations from EtOH gave the analytical sample: mp 186.5–187.5 °C; $[\alpha]^{25}D$ −113.7° (*c* 1.0, $CHCl_3$); NMR (Me_2SO-d_6) δ 4.72 (s, 2, CH_2), 6.18 (s, 1, $H_{1'}$).

Anal. Calcd for $C_{29}H_{24}N_2O_9S$ (576.580): C, 60.41; H, 4.20; N, 4.86; S, 5.56. Found: C, 60.63; H, 4.49; N, 5.03; S, 5.60.

2-(2,3,5-Tri-*O*-benzoyl-β-D-ribofuranosyl)-5-dimethylamino-6*H*-1,2,4-oxadiazin-3(2*H*)-one (13). A. A solution of 0.430 g (3.0 mmol) of **5**, 30 mg of ammonium sulfate, and 4.3 ml of HMDS in 37 ml of dry dioxane was refluxed for 15 h. After cooling, dioxane and HMDS were removed in vacuo and the residue was then dried at the vacuum pump for 1 h. The semisolid residue was taken up in 30 ml of dry 1,2-dichloroethane after which 3 g of 4A molecular sieves and 1.526 g (3.0 mmol) of 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-β-D-ribofuranose were added. The flask was then flushed with nitrogen and stoppered with a rubber septum. After $SnCl_4$ (0.4 ml, 3.0 mmol) was added, the reaction mixture was stirred at room temperature for 18 h. The reaction mixture was then poured into 15 ml of saturated aqueous $NaHCO_3$. After stirring well, Celite was added followed by filtration through a Celite pad. After the filter cake was washed well with CH_2Cl_2, the organic phase was washed with 25 ml of saturated aqueous NaCl solution and then dried over Na_2SO_4. Removal of the solvent in vacuo gave 1.627 g of solid residue. Dry column chromatography on 100 g of dry column silica gel, eluting with 150 ml of 99:5 CH_2Cl_2–MeOH, gave 1.081 g (61.3%) of white solid. Two recrystallizations from EtOH gave the analytical sample: mp 185–186 °C; $[\alpha]^{25}D$ −56.4° (*c* 1.0, $CHCl_3$); NMR (Me_2SO-d_6) δ 2.94, 3.19 (s, 6, NMe_2), 4.63 (s, 2, CH_2), 6.44 (d, J = 5 Hz, 1, H_1).

Anal. Calcd for $C_{31}H_{29}N_3O_9$ (587.585): C, 63.37; H, 4.97; N, 7.15. Found: C, 63.19; H, 4.67; N, 7.17.

B. Dimethylamine was bubbled into a solution of 0.179 g (0.3 mmol) of **12** in 7 ml of dry dioxane for 5 min. After stirring at room temperature for 12 min longer, dioxane was removed in vacuo. Preparative TLC of the residue on silica gel, eluting with 95:5 $CHCl_3$–MeOH, gave 0.094 g (53.3%) of a white solid, which was the same as the product obtained in A by comparison of TLC, melting point, ir, and NMR.

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Registry No.—**1**, 5766-95-0; **1** anion, 59696-54-7; **2**, 59696-55-8; **3**, 59696-56-9; **4**, 5767-01-1; **5**, 59696-57-0; **8a**, 59696-58-1; **8b**, 59696-59-2; **8c**, 59696-60-5; **10**, 5767-08-8; **11**, 5767-15-7; **12**, 59696-61-6; **13**, 59696-62-7; P_2S_5, 1314-80-3; dimethylamine, 124-40-3; 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-β-D-ribofuranose, 6974-32-9; 1,2,3,5-tetra-*O*-acetyl-β-D-ribofuranose, 13035-61-5.

References and Notes

(1) H. Kornowski, M. Trichot, B. Delage, and M. Phan-Chi-Don, *Bull. Soc. Chim. Fr.*, 679 (1966).
(2) R. Masingale, S. R. Bryant, C. G. Skinner, J. Nash, and P. F. Kruse, Jr., *J. Med. Chem.*, **12**, 152 (1969).
(3) V. Skaric, B. Gaspert, I. Jerkunica, and D. Skaric, *Croat. Chem. Acta*, **37**, 199 (1965).
(4) D. M. Brown and M. J. E. Hewlings, *J. Chem. Soc. C*, 2050 (1968).
(5) H. Kornowski, M. Trichot, and B. Delage, *Bull. Soc. Chim. Fr.*, 683 (1966).
(6) H. Vorbruggen, *Angew. Chem., Int. Ed. Engl.*, **11**, 305 (1972).
(7) H. Vorbruggen, K. Krolikiewicz, and U. Niedballa, *Justus Liebigs Ann. Chem.*, 988 (1975).
(8) A. J. Jones, D. M. Grant, M. W. Winkley, and R. K. Robins, *J. Am. Chem. Soc.*, **92**, 4079 (1970).
(9) U. Niedballa and H. Vorbruggen, *J. Org. Chem.*, **39**, 3654 (1974).
(10) L. B. Townsend in "Synthetic Procedures in Nucleic Acid Chemistry", Vol. 2, W. W. Zorbach and R. S. Tipson, Ed., Wiley-Interscience, New York, N.Y., 1973, p 267.
(11) K. A. Watanabe and J. J. Fox, *J. Heterocycl. Chem.*, **6**, 109 (1969).
(12) G. E. Hilbert and T. B. Johnson, *J. Am. Chem. Soc.*, **52**, 4489 (1930).
(13) P. Dea, G. R. Revankar, R. L. Tolman, R. K. Robins, and M. P. Schweizer, *J. Org. Chem.*, **39**, 3226 (1974).
(14) L. F. Johnson and W. C. Jankowski, "Carbon-13 NMR Spectra", Wiley, New York, N.Y., 1972, p 170.
(15) R. A. Long and L. B. Townsend, *Chem. Commun.*, 1087 (1970).
(16) (a) V. Skaric, B. Gaspert, M. Hohnjec, and G. Lacan, *J. Chem. Soc., Perkin Trans. 1*, 267 (1974); (b) V. Skaric, B. Gaspert, and M. Hohnjec, *J. Chem. Soc. C*, 2444 (1970).
(17) V. Skaric and B. Gaspert, *J. Chem. Soc. C*, 2631 (1969).