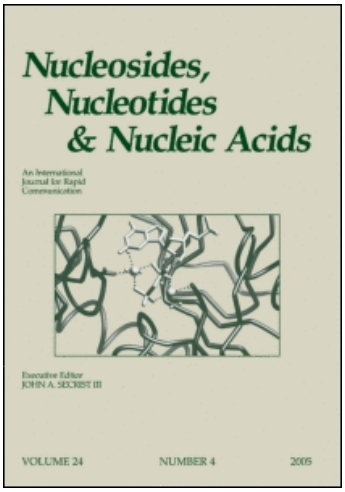

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SYNTHESIS OF 2-S-DIOXO ISOSTERES OF PURINE AND PYRIMIDINE NUCLEOSIDES. V. ^{13}CDNMR STUDY OF *SYN-ANTI* EQUILIBRIUM IN A GLUCOSIDE OF A N-SUBSTITUTED BARBITURIC ACID ISOSTERE

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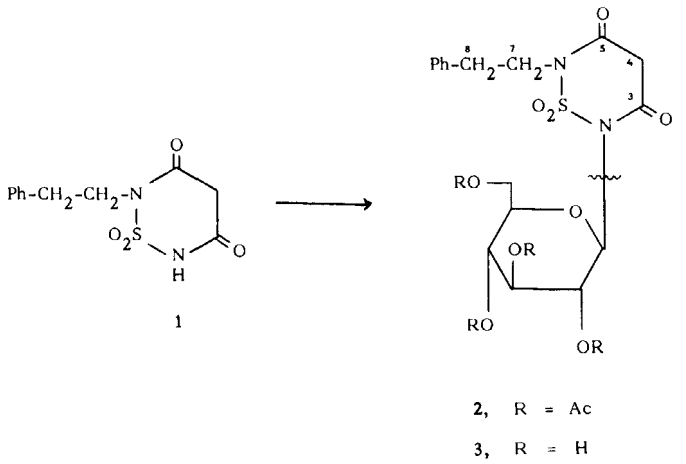
Abstract. *Glycosylation of 2-(2-phenylethyl)-1,2,6-thiadiazin-3,5-dione 1,1-dioxide (**1**) **via** the "silyl method" is described. The reaction favours **N**-substitution and the tetra-O-acetyl-β-D-glucopyranoside **2** as well as the free nucleoside **3** have been isolated and characterized by UV and NMR studies. Nucleoside **2** exists as rotationally restricted **syn-anti** conformers at room temperature. From ^{13}CDNMR studies over the temperature range 234-333 K the enthalpy ΔH^{++} and entropy ΔS^{++} of activation has been calculated.*

Continuing with our studies on glycosylation reactions of 1,2,6-thiadiazine 1,1-dioxide derivatives[1-5], the results on the synthesis of glucosyl derivatives of 2-(2-phenylethyl)-1,2,6-thiadiazin-3,5-dione 1,1-dioxide (**1**)[6] is now described.

Compound **1** can be regarded as a sulfur dioxi analog of 1-substituted barbituric acid derivatives. A few reports on the synthesis of glycosyl derivatives of barbituric acid have been described. Most of them[7,8] deal with transformations of other pyrimidine nucleosides into barbituric acid derivative nucleosides. One case of direct **N**-glycosylation of barbituric acid by reaction of trimethylsilyl derivative of the base and tri-O-acetyl-D-ribofuranose bromide has been reported[9]. In our case, glycosylation of **1** using the "silyl method", in the presence of Friedel-Crafts catalysts[10], gave good results. Thus, reaction of the trimethylsilyl derivative of **1** with 1,2,3,4,6-penta-O-acetyl-β-D-glucopyranose afforded one nucleoside (**2**), as a glass, in a high yield. The site of glycosyl-



SCHEME 1

ation was determined by comparing its UV spectra with those of N,N'-di-substituted 1,2,6-thiadiazin-3,5-dione 1,1-dioxides. The similarity of the UV data in all the cases and the chemical shift of the anomeric proton are only consistent with an N-nucleoside.

The ^{1}H-NMR spectrum of **2** in $CDCl_3$ shows some signal broadened. When the spectrum was registered at 323 K these signals became narrow and the signal corresponding to H-2' could be seen as a triplet. On cooling this sample to 293 K the original spectrum gradually reappeared. These facts indicate that a dynamic process takes place on heating. In order to discard the possibility of a tautomeric equilibrium which is usual in these heterocycles[11], drops of TFA were added to the sample and the spectrum registered once more. No change was observed in the spectrum and thus the existence of a mixture of two rotational isomers of **2** (*syn* and *anti*) due to restricted rotation about the glycosidic bond seems more probably.

In order to study the problem variable temperature ^{1}H-NMR experiments of **2** were carried out using $CDCl_3$ and DMSO-d_6 as solvents. The data found are gathered in Table 1.

The ^{1}H-NMR spectrum in $CDCl_3$ at 333 K shows the average signal of anomeric *syn* and *anti* protons as a broad multiplet and the other signals as well resolved multiplets. The more deshielded signal corresponds to H-2' and it appears as a triplet as well as the corresponding ones of H-3' and H-4' with coupling constants ($J \simeq 9.5$ Hz) according to the $^4C_1$ conformation. The two H-4 protons of the thiadiazine ring appear as an AB system with a geminal coupling constant of -18.1 Hz. This fact is different to what occurs in the ^{1}H-NMR spectrum of **1** in which the two H-4 appear as a singlet[6].

TABLE 1. ^{1}H-NMR chemical shifts[a] (δ, ppm) and coupling constants (J, Hz) of compound **2**

Temp. (K)	Solvent	H-1'	H-2'	H-3'	H-4'	H-5'	H-6', H-6"	H-4(A,B)	H-7	H-8
293	$CDCl_3$	-	5.78(b.m.)	5.22(t) ^3J = 9.3	5.12(t) ^3J = 9.8	3.77(d.t.) $^3J_{4',5'}$=10.0 $^3J_{5',6'}$=3.3	≃ 4.1(b.m.)	≃ 4.0(b.m.)	4.0(b.m.)	2.88(t) $^3J_{8,7}$=7.8
333	$CDCl_3$	5.56(b.m.)	5.83(t) ^3J = 9.2	5.26(t) ^3J = 9.4	5.16(t) ^3J = 9.7	3.79(m)	4.21(m)	4.06(d) 3.79(d) ^2J = -18.1	4.07(t) ^3J = 7.8	2.96(t) ^3J = 7.8
295	DMSO-d_6	6.24(d)(anti) ^3J = 9.4 5.81(d)(syn) ^3J = 9.2	5.64(t) ^3J = 9.9	5.54(t) ^3J = 9.4	4.9(b.m.)	4.35(b.m.)	4.1(b.m.)	≃ 3.9(b.m.)	≃ 3.9(b.m.)	2.87(t) ^3J = 7.5
333	DMSO-d_6	6.0(b.m.)	5.6(b.m.)	5.50(t)	4.90(t)	4.30(d.t.)	4.1(b.m.)	4.0(b.m.)	4.0(b.m.)	2.87(t)
353	DMSO-d_6	5.9(b.m.)	5.65(t) ^3J = 9.0	5.49(t) ^3J = 9.5	4.93(t) ^3J = 9.8	4.29(d.t.) $^3J_{4',5'}$=10.1 $^3J_{5',6'}$=3.5	4.1(m)	≃ 4.0(b.m.)	3.99(t) ^3J = 7.6	2.88(t) ^3J = 7.6

a) for numbering see Scheme 1.

In the spectrum at 293 K ($CDCl_3$) the anomeric protons signals could not be observed. However, the ^{1}H-NMR spectrum of **2** in DMSO-d_6 at 295 K shows two doublets corresponding to the anomeric protons of the *syn* and the *anti* conformers with coupling constants of 9.2 and 9.4 Hz respectively indicating a β-anomer. As usual[12], the more deshielded signal was assigned to the *anti* rotamer. The coalescence temperature for the anomeric protons signals in DMSO is between 323 K and 333 K. In this solvent, the anomeric signals appear more deshielded than those corresponding to H-2' as expected. The other difference with respect to the spectra in $CDCl_3$ is that the signal belonging to the H-4 protons of the heterocycle appears as a broad multiplet at all temperatures.

The signals affected by the increase in temperature are those corresponding to H-1', H-2', H-6', H-6", of the sugar moiety, H-7 of phenylethyl substituent and H-4 of the heterocycle.

X-ray studies of 2,6-disubstituted 1,2,6-thiadiazin-3,5-dione 1,1-dioxides[13] show for these compound, the boat conformation with the C-4 and S atoms at the flaps and the N-substituents at the same side of the plaine, probably the same occurs in nucleoside **2** although it is not possible to study the compound in the solid state.

The ^{13}C-NMR study of **2** was carried out in $CDCl_3$ and consisted in experiments between 234 K and 333 K temperatures. The ^{13}C-NMR spectrum at 234 K shows almost all signals split due to the existence of *syn* and *anti* rotamers whilst the spectrum at 333 K shows all average signals *(see Fig. 1)*. The ^{13}C-NMR data of spectra at 234 and 333 K are gathered in Table 2.

The assignments were made by chemical correlation[6,15] and $^1J_{C\text{-}n,H\text{-}n}$ measurements[16]. Differentiation between C-3 and C-5 was easy since C-3 next to the glycosidic bond is more affected by temperature variation. The signals of the *anti* rotamer were tentatively assigned to those more deshielded but the assigment may be interchanged.

Nuclear magnetic resonance has been extensively used for determination of thermodynamic parameters associated with dynamic processes. Most studies have utilized ^{1}H-NMR[12,17] or ^{19}F-NMR[18], but ^{13}C-NMR[19] has proven to be a more useful technique to determine rate constants from the line width and the coalescence temperature studies. In this case ^{13}C-DNMR data over the temperature range 264 -328 K were used to study the kinetic process associated with rotational conversion between *syn* and *anti* conformers of nucleoside **2**.

The rate constants were determined by the method of line shapes[20]. Table 3 contains the spectral parameters and rate constants using the anomeric carbon signals.

TABLE 2. ^{13}C-NMR chemical shifts (ppm) and coupling constants (Hz) of **2** at 234 and 333 K[a)]

Temp.	234 K	333 K
C-3 *(anti)*	162.5	162.0 (b.s) -
C-3 *(syn)*	161.1	
C-4 *(anti)*	45.2	44.0 (t), ^{1}J = 136.0
C-4 *(syn)*	44.1	
C-5 *(anti)*	161.8	161.8 (t.t.), $^{2}J^{*}$ = |8.1|, $^{3}J^{*}$ = 2.4
C-5 *(syn)*	161.1	
C-7 *(anti)*	46.6	46.3 (t), ^{1}J = 146.1
C-7 *(syn)*	46.2	
C-8 *(anti)*	35.0	35.2 (t), ^{1}J = 131.8
C-8 *(syn)*	34.9	
C-*i* *(anti)*	136.7	137.0 -
C-*i* *(syn)*	136.3	
C-*o*	128.6	128.8 (d), ^{1}J = 170.2
C-*m*	128.9	129.0 (d), ^{1}J = 168.8
C-*p* *(anti)*	127.1	127.1 (d), ^{1}J = 160.0
C-*p* *(syn)*	126.9	
C-1' *(anti)*	82.3	82.3 (d), ^{1}J = 151.1
C-1' *(syn)*	80.4	
C-2' *(anti)*	68.4	68.8 (d), ^{1}J = 154.0
C-2' *(syn)*	67.1	
C-3'	74.3	75.3 (d), ^{1}J = 140.6
C-4'	66.7	67.9 (d), ^{1}J = 154.6
C-5' *(anti)*	72.5	73.4 (d), ^{1}J = 150.1
C-5' *(syn)*	72.3	
C-6' *(anti)*	61.2	61.6 (d), ^{1}J = 148.0
C-6' *(syn)*	60.7	

a) $CDCl_3$ as solvent

* The ^{2}J and ^{3}J values may be reversed[14)].

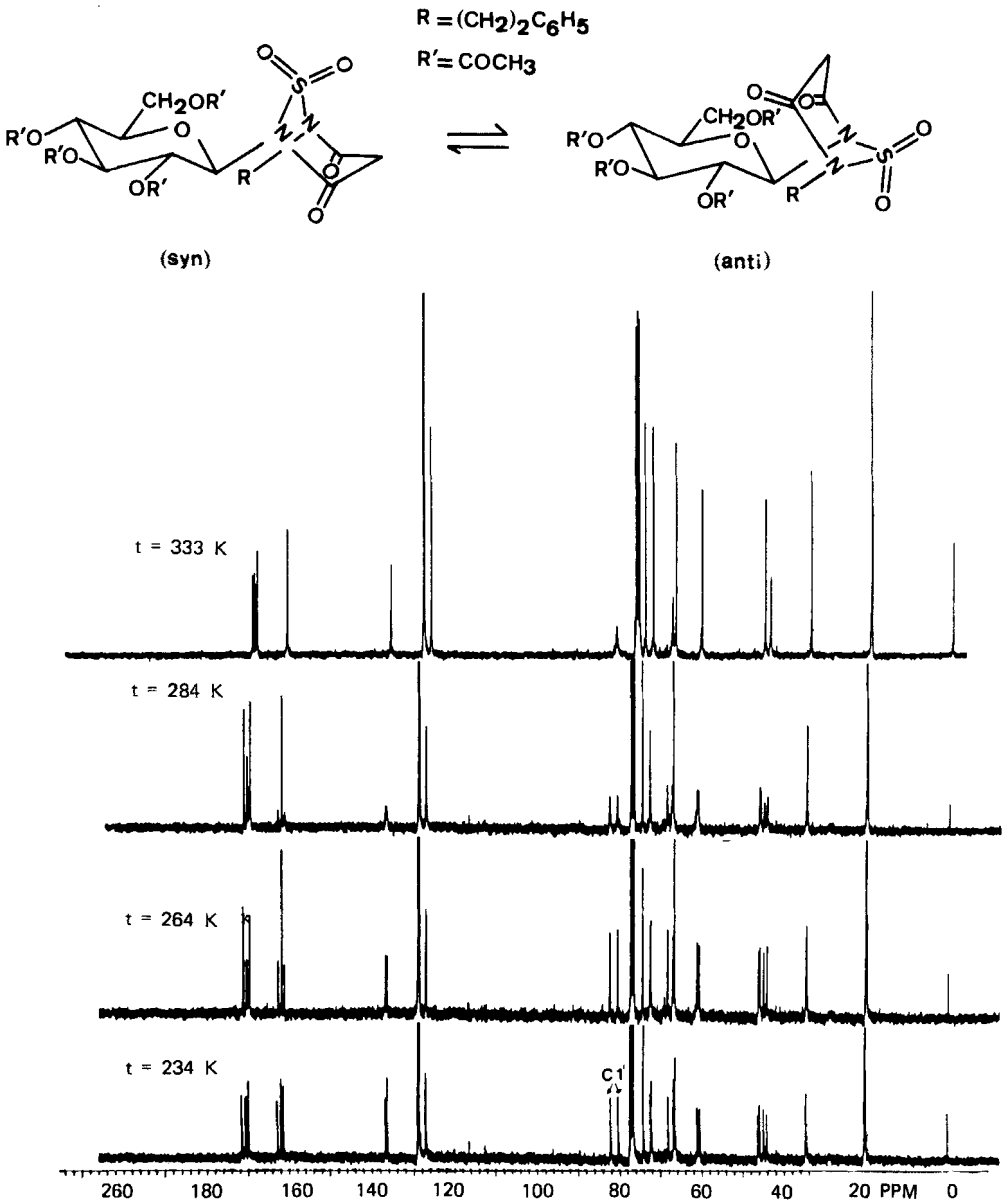


FIGURE 1

TABLE 3. Spectral parameters and rate constants $(K_r)^{a)}$

Temp. (K)	$\Delta\nu^{b)}$	K_r (sec^{-1})
264	3.22	10.116
274	6.18	19.415
284	11.05	34.714
298	52.96	160.374
308	58.07	182.432
318	83.20	370.043
328	31.25	985.203

a) ν_{AB}, the difference of chemical shifts in hertz (140 Hz) between anomeric carbons of *anti* and *syn* rotamers, has been measured in the ^{13}C-NMR spectrum at 234 K. The transmission coefficient value used has been unity.

b) $\Delta\nu$ is the difference between line width of anomeric carbon and line width of TMS used as reference.

From the Eyring equation of absolute reaction rate theory[21] the enthalpy ΔH^{++} and entropy ΔS^{++} of activation were calculated by least-squares linear regression analysis (r^2 = 0.98). The values of ΔH^{++}, 11.4 kcal/mol, and ΔG^{++}, 14.8 kcal/mol found, at coalescence temperature (313 ± K), indicate a lower rotational energy barrier than those described for pteridine deblocked glucoside derivatives[17]. This is probably due to the boat conformation of thiadiazine 1,1-dioxide ring since the SO_2 group outside the plane of glycosidic bond produces a lower steric hindrance than the CO group in the pteridine ring. The value of ΔS^{++} found is -10.6 e.u. Slight negative activation entropies are often found in DNMR studies of intramolecular process in which alkyl groups are squeezed past each other, or past other obstructing groups, in the transition state. It seems reasonable to assume that in this case the negative entropy is due to decreased rotational freedom of N-substituents of **2** in the transition state.

Calculations were made taking into account that the process may be considered as a degenerate process since population of *syn* and *anti* conformers are almost equal as can be meaured from the anomeric proton signal integrals in the spectrum in DMSO-d_6 at 295 K.

Removal of acetyl protecting groups of nucleoside **2** with ethanolic ammonia solution yields the deblocked nucleoside **3**. Its ^{1}H-NMR spectrum in DMSO-d_6 + D_2O shows a mean signal for the anomeric protons. This fact indicates that the rotation about the glycosidic bond is not restricted at room temperature in this nucleoside. The signals belonging to H-4', H-4 and H-7 cannot be seen since they appear overlapped with DMSO-d_6 and D_2O signals.

It can be concluded that the slight restricted rotation about the glycosidic bond at room temperature found in nucleoside **2** disappears when the steric hindrance is lower as in the case of deblocked glucose moiety of **3**.

Recently, a facile synthesis of 1,3-dimethyl barbituric acid C-nucleosides has been described[22], which deal with the reaction between a deprocted sugar and **N,N'**-dimethylbarbituric acid in alkaline medium. Attempts to extend this procedure to obtain C-nucleosides of N-substituted 1,2,6-thiadiazin-3,5-dione 1,1-dioxide derivatives failed although several thiadiazin-3,5-dione derivatives with different pK_a values were used as starting materials.

EXPERIMENTAL

Ultraviolet spectra were measured on a Perkin-Elmer 550 spectrophotometer. ^{1}H-NMR were recorded on a Varian XL-300 instrument operating at 300 MHz, using $CDCl_3$ or DMSO-d_6 as solvents and TMS as internal standard. Two dimensional scalar shift-correlated ^{1}H-NMR spectra were recorded in the same spectrometer using 90°-t_1-90° pulse sequence referred to as COSY. The following parameters were used: number of increments, 512; 90° pulse width, 13.7 μs; relaxation delay, 2s; sweep width in t_1 and t_2, 1500 Hz and 1024 x 1024 transformed data points.

^{13}C-NMR decoupled and coupled spectra were measured on a Varian XL-300 instrument operating at 75 MHz, using $CDCl_3$ as solvent and TMS as internal standard. The decoupled spectra were recorded at twelve different temperatures in the range 234 -333 K.

Column chromatography was performed on Merck silica gel 60 (230-400 mesh), and preparative thin coated with 2 mm layer of silica gel PF_{254} (Merck).

Compounds were detected with UV light (254 nm) or by spraying the plate with ethanol:sulphuric acid (3:1) and heating.

6-Phenylethyl-2-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-1,2,6-thiadiazin-3,5-dione 1,1-dioxide (**2**).

To a stirred solution of 0.97 g (0.25 mmole) of 1,2,3,4,6-penta-O-acetyl-β-D-glucopyranose in 50 ml of dry methylene chloride, a solution of the silyl derivative of **1** (prepared from **1** (0.67 g, 0.25 mmole) and hexamethyldisilazane (15 ml) in the presence of $(NH_4)_2SO_4$ as catalyst, under N_2 atmosphere) in methylene chloride was added. The mixture was treated with 3 ml of boron trifluoride etherate and stirred for four hours at room temperature with exclusion of humidity. The reaction mixture was then treated with saturated sodium hydrogen carbonate solution (100 ml). The organic phase was separated, dried over sodium sulphate and evaporated under reduced pressure. The residue (1.4 g) was chromatographed on silica gel column using chloroform: methanol (30:1) as eluent. The oily residue was rechromatographed using preparative TLC eluting with chloroform-methanol (40:1). The higher running band was separated and purified on silica gel column, affording 1.2 g (79% yield) of the β-anomer **2**, obtained as a white glass.

UV (MeOH) : 271.5 nm (log ε = 2.8).

Anal. Calcd. for $C_{25}H_{30}N_2O_{13}S$: C, 50.16; H, 5.05; N, 4.68; S, 5.35. Found: C, 50.34; H, 5.12; N, 4.75; S, 5.05.

6-Phenylethyl-2-(β-D-glucopyranosyl)-1,2,6-thiadiazin-3,5-dione 1,1-dioxide (**3**).

A solution of 0.3 g (1.1. mmole) of **2** in 10 ml of saturated methanolic ammonia was stirred at room temperature for 6 hours. The solution was evaporated to dryness, and the residue was purified by TLC using chloroform : methanol (3 : 1) as eluent. The lower running band afforded 187 mg (89% yield) as a white powder.

^{1}H-NMR (DMSO-d_6 + D_2O) δ: 7.22-7.25 (m, 5H, C_6H_5); 4.29 (d, 1 H, $J_{1',2'}$ = 8.6 Hz, H-1'); 3.77 (dd, 1 H, $J_{3',2'}$ = 6.8 Hz, $J_{3',4'}$ = 9.9 Hz, H-3'); 3.76 (dd, 1H, H-2'); 3.15 (m, 1H, H-5'); 3.05 (t, 2 H, $J_{8,7}$ = 7.9 Hz, H-8); 2.8 (m, 2 H, H-6').

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