Rotational Isomerism in 6-β-D-Glucopyranosides of Methyl-1,2,6-thiadiazin-3(2*H*)-one 1,1-dioxides

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Glucopyranosides of methyl-1,2,6-thiadiazin-3(2*H*)-one 1,1-dioxides have been synthesized and their rotational isomerism studied. Two of them exist, in solution, as mixtures of *syn*:*anti* rotamers, at room temperature. In the case of the diglucoside, the barriers to rotation about both glycosidic bonds have been calculated by dynamic ^{13}C NMR.

Glycosyl derivatives of 1,2,6-thiadiazine 1,1-dioxides, synthesized as SO_2 analogues of nucleosides have been the subject of previous studies.[1] Some of them, particularly when the sugar moiety is a glucopyranose, have shown interesting conformational problems regarding the relative position of the base and the sugar moieties about the glycosidic bond.[2,3] Since it is well-known that *syn*:*anti* equilibrium in pyrimidine nucleosides is strongly affected by the presence of substituents at the 6-position,[4] we decided to study the β-D-glucopyranosides of the SO_2 analogue of 6-methyluracil, namely 5-methyl-2*H*-1,2,6-thiadiazin-3(2*H*)-one (**1**).[5]

Results and Discussion

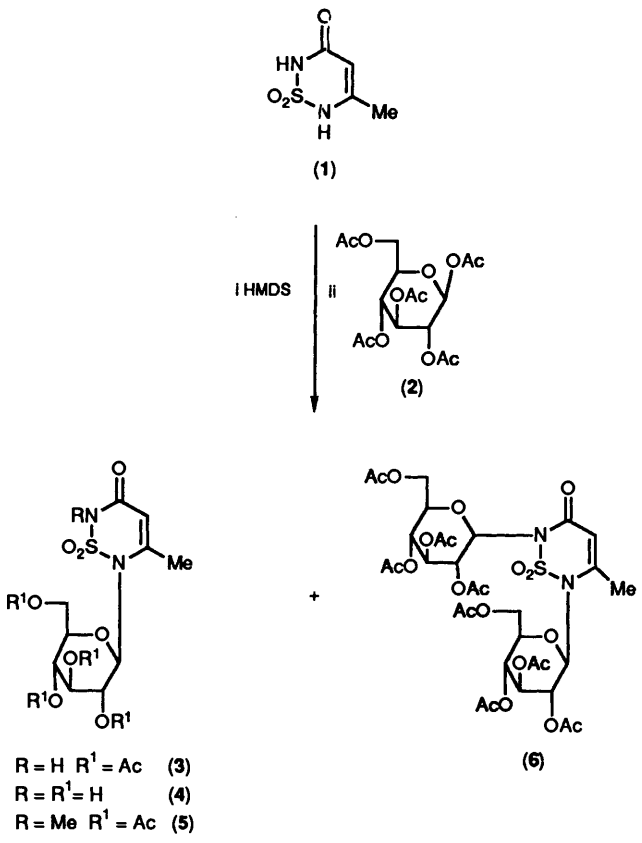
Synthesis.—The glycosylation method chosen was the 'silyl procedure',[6] and thus, thiadiazine (**1**) was first silylated with HMDS under a nitrogen atmosphere. Reaction of this silyl derivative with 1,2,3,4,6-penta-*O*-acetyl-β-D-glucopyranose (**2**), in dichloromethane, with $Et_2O{\cdot}BF_3$ as catalyst, afforded a complex mixture from which it was possible to isolate the mono- and di-glucopyranosides (**3**) and (**6**) respectively, (Scheme 1).

Treatment of (**3**) with methanolic ammonia afforded the fully deprotected glucoside (**4**). The *N*-methyl derivative (**5**) was obtained from (**3**) and methyl iodide.

Structural Assignments.—The structures of the compounds were established according to analytical and NMR data (Tables 1 and 2). In the ^{13}C NMR spectrum ([$^2H_6$]DMSO) of (**3**) the signal of the anomeric carbon appeared at 82.5 ppm, consistent with an *N*-glucoside. The site of glycosylation was definitively established as N(6) and not N(2), by ^1H NOE difference experiments. Irradiation of the doublet of the anomeric proton (δ 5.62) showed an NOE effect (8%) on the singlet of the 5-methyl group (δ 2.11). Similar NOEs were observed for the deprotected glucoside (**4**) and the methyl derivative (**5**). In the latter, the ^{13}C NMR signal of the methyl group at 33.6 ppm clearly ruled out the possibility of an *O*-substitution having taken place.

The other nucleoside isolated from the glycosylation mixture was identified as the *N*(2),*N*(6)-diglucosyl derivative (**6**) on the basis of NMR data: the sugar moieties had to be at N(6) [similar ^1H NOE as in (**3**), (**4**), and (**5**)] and at N(2) (chemical shift of the other anomeric proton, δ 5.71 and carbon, 85.2 ppm).

In all cases, the anomeric configuration for the glucosides was established as β according to the values of the proton $J_{1',2'}$ coupling constants which range from 9.0 to 9.5 Hz consistent with a *trans* diaxial disposition in the expected $^4C_1$ conformation.[7]



Scheme 1.

Finally, the UV data of all the glucosides are in agreement with the structures proposed as deduced from comparison of the spectra with those of suitable *N*-alkyl models.[8]

Conformational Studies.—Valuable information concerning the *syn*:*anti* equilibrium about the glycosidic bond of nucleosides can be gained from NMR.[9] Thus, there are no indications in the spectra of (**3**) and (**4**) of the existence of a restricted rotational equilibrium at room temperature. The fact that the spectra ($CDCl_3$) of (**3**) showed very broad signals, which did not collapse on heating, can be explained on the basis of a prototropic exchange occurring at the NH–CO moiety.

Table 1. ^{1}H NMR parameters: chemical shifts[a] (ppm) and coupling constants (Hz) of glucopyranosides (**3**)–(**6**).

Compound	H(1′)	H(2′)	H(3′)	H(4′)	H(5′)	H(6′a)	H(6′b)	H(4)	Me(5)
(**3**)[b]	5.62(d)	5.36(t)	5.15(t)	4.99(t)	———	4.13–4.04(m)	———	5.11(d)	2.11(d)
		$J_{1'2'}$ 9.5	$J_{2'3'}$ 9.6	$J_{3'4'}$ 9.3	$J_{4'5'}$ 9.3				$J_{4,Me}$ 0.9
(**4**)[c]	5.30(d)	4.11(t)	———			3.66–3.08(m)	———	4.75(s)	2.10(s)
		$J_{1'2'}$ 9.0							
(**5**)[d]	———5.89(br s)———		5.28(t)	5.21(t)	3.84(m)	4.22(q)	4.30(q)	5.65(d)	2.02(d)
				$J_{3'4'}$ 9.2	$J_{4'5'}$ 9.6	$J_{5'6'a}$ 2.2	$J_{5'6'b}$ 4.4		$J_{4,Me}$ 1.0
						$J_{6'a6'b}$ −12.8			
(**6**)[e] N-2	5.71(d)	5.82(t)	5.39*(t)	5.08(t)					
		$J_{1'2'}$ 9.3	$J_{2'3'}$ 9.3	$J_{3'4'}$ 9.2					
					———	4.21–4.06(m)	———	5.96(d)	2.30(d)
									$J_{4,Me}$ 1.1
N-6	5.63(d)	5.31(t)	5.38*(t)	4.96(t)					
		$J_{1'2'}$ 9.2	$J_{2'3'}$ 9.3	$J_{3'4'}$ 9.3					

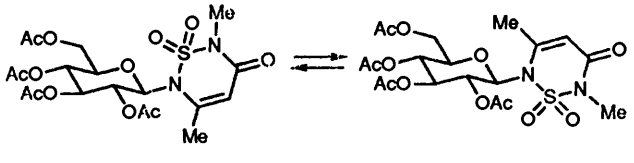
[a] δ(TMS) = 0. [b] [$^2H_6$]DMSO at 293 K. [c] [$^2H_6$]DMSO + D_2O at 293 K. [d] $CDCl_3$ at 328 K, δ_{N-Me} = 3.33. [e] [$^2H_6$]DMSO at 393 K.

Table 2. ^{13}C NMR chemical shifts[a] of glucopyranosides (**3**)–(**6**).

Carbon	(**3**)[b]	(**4**)[c]	(**5**)[d,e]	(**6**)[d]	(**6a**)[f]	(**6b**)[f]	(**6c**)[f]	(**6d**)[f]
C(3)	159.5	165.1	160.9	n.o.	161.9		159.4	
C(4)	107.7	91.2	106.1	114.6	114.6, 114.5[g]			
C(5)	145.7	151.9	152.2	n.o.	147.5	150.5	147.5	150.5
Me(5)	20.3	24.2	20.4	20.6	21.9	21.6	21.9	21.6
C(1′)	82.5	81.9	81.3	81.3	82.5	79.2	82.5	79.2
C(2′)	69.5	69.8	68.9	69.3	69.1	69.8	69.1	69.8
C(3′)	72.9	79.4	74.9	75.2	74.5	74.3	74.5	74.3
C(4′)	67.6	69.6	68.0	68.0	66.9	66.7	66.9	66.7
C(5′)	72.6	77.5	73.6	74.0	73.1	73.6	73.1	73.6
C(6′)	61.5	60.8	61.6	61.5	61.1	60.5	61.1	60.5
C(1″)	—	—	—	85.2	85.0		84.2	
C(2″)	—	—	—	68.3	67.8		68.2	
C(3″)	—	—	—	75.2	74.5		74.5	
C(4″)	—	—	—	68.0	67.0		67.0	
C(5″)	—	—	—	73.6	72.8		73.0	
C(6″)	—	—	—	61.6	61.1		61.1	

[a] δ(TMS) = 0. [b] [$^2H_6$]DMSO at 293 K. [c] [$^2H_6$]DMSO + D_2O at 293 K. [d] $CDCl_3$ at 328 K. [e] δN-Me = 33.6. [f] $CDCl_3$ at 246 K. [g] Signals not assigned.

The ^{1}H NMR spectrum ($CDCl_3$) at room temperature (293 K) of (**5**) showed significant peak broadening for the H(5′), H(6′a), and H(6′b) protons and double signals for H(1′) and H(2′). Particularly, the anomeric proton appeared as two broad signals (δ_{syn} 5.8, δ_{anti} 6.0). These results are consistent with the existence of (**5**) as a mixture of two rotational isomers, in solution, at room temperature, (Scheme 2).



syn (**5**) *anti*

Scheme 2. The *syn* and *anti* ranges of the glycosidic bond have been defined by analogy with those used in the pyrimidine nucleosides.[10]

The rest of the protons less affected by the dynamic process showed the expected multiplicity and thus, it was possible to assign, at 328 K (Table 1), all the signals and measure the coupling constants. The fact that the H(1′) and H(2′) signals did not collapse at 328 K can be explained by taking into account that these protons are the most affected by the dynamic process since they lie close to the SO_2 and methyl groups, responsible for the restricted rotation (Scheme 2).

The ^{13}C NMR spectrum ($CDCl_3$) of (**5**) at 293 K also showed broad signals for the carbons of the heterocyclic moiety, for C(1′) and C(2′) and to a lesser extent, for C(5′) and C(6′). At 328 K only the latter became sharp in agreement with the ^{1}H NMR data.

More interesting is the case of the diglucoside (**6**). The ^{1}H NMR spectra of (**6**), in $CDCl_3$ and [$^2H_6$]DMSO, at 293 K were very complex and only in [$^2H_6$]DMSO, at 393 K, was it possible to assign all the signals by means of a COSY homonuclear experiment[11] (Table 1).

The ^{1}H NMR spectrum ($CDCl_3$) of (**6**), at 293 K, showed two broad signals for each of the anomeric protons. This might be due either to two hindered rotations around the C–N bonds (resulting in four different rotamers as outlined in Scheme 3), or to the fact that the existence of two rotamers on one sugar could bring about significant changes in the chemical shifts of the other glycoside. This point had to be clarified first and so dynamic ^{13}C NMR was chosen as the most suitable means to solve this problem.

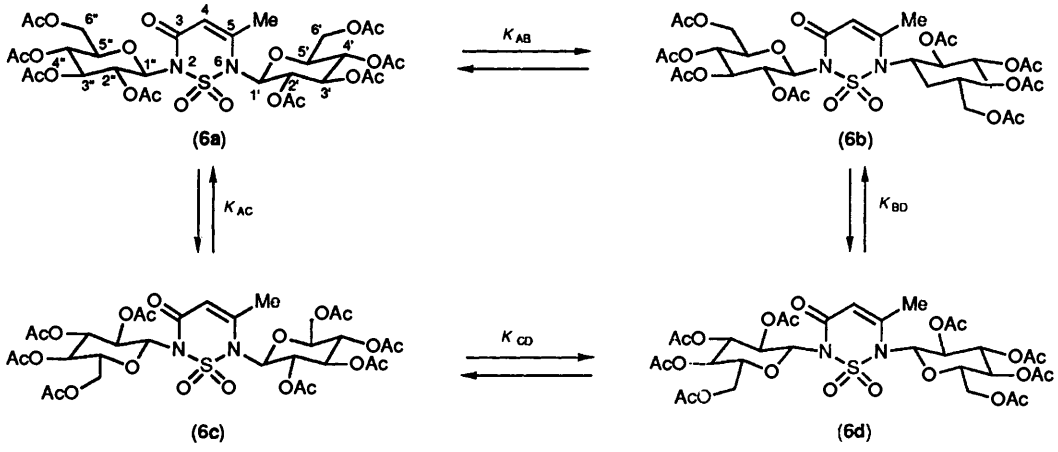
At low temperature, 246 K, the ^{13}C NMR spectrum ($CDCl_3$) showed double signals for all the carbons. If rotation around both bonds were not restricted, then either C(3) or C(5) (the carbon atom of the base lying closer to the sugar involved) should be more influenced by the dynamic process. However, the spectrum showed two bands for each one of them with a similar difference in chemical shift while C(4) had a much smaller one. This fact suggested that interconversion of the four rotamers depicted in Scheme 3, was most likely taking place.

This means that for each anomeric carbon, four bands corresponding to forms (**6a–d**) are to be expected, at low temperature. However, at 246 K, only two bands were observed for each anomeric carbon. This can be explained by assuming that rotation around one C–N bond does not significantly alter the chemical shifts of the carbons of the sugar at the other N. In other words, the chemical shift of C(1′) would be the same in (**6a**) and (**6c**), and that of C(1″) the same in (**6a**) and (**6b**). On the basis of this hypothesis, it would be possible to measure, independently, the two barriers to rotation, equations (1) and (2).

$$k_{AC} = k_{BD}[\text{rotation around C–N(2)}] \quad (1)$$

$$k_{AB} = k_{CD}[\text{rotation around C–N(6)}] \quad (2)$$

Therefore, a dynamic ^{13}C NMR study was carried out, in the



Scheme 3.

Table 3. Rate constants calculated in the anomeric carbon signals of (**6**).

T/K	$k_{85.0}$/s^{-1}	$k_{84.2}$/s^{-1}	$k_{82.5}$/s^{-1}	$k_{79.1}$/s^{-1}
256	2.95	1.66	—[a]	—[a]
266	6.03	3.30	—[a]	—[a]
271	8.65	5.25	1.10	2.89
276	13.10	8.39	1.73	6.44
281	17.34	11.47	8.32	11.84
286	20.48	15.77	9.11	12.25
292	31.10	25.13	24.35	25.60
298	45.71	38.96	30.32	37.85
303	62.52	54.66	73.51	75.46
308	80.03		120.17	124.09
313	88.57		163.52	151.74
318	141.45		194.93	206.72
323	196.53		—[b]	—[b]
328	214.81		—[b]	—[b]

[a] No significant variations are observed in the line widths. [b] Not calculated due to the lack of the signal:noise ratio.

range 246–328 K, for both anomeric carbons. On raising the temperature, the two signals at 85.0 and 84.2 ppm collapsed to one (85.2 ppm) at 308 K, whereas the couple at 82.5 and 79.2 ppm reached coalescence at 328 K. In both cases, a downfield shift could be observed at higher temperatures. Since the average signal of the anomeric carbon of the model compound (**5**) appeared at 81.3 ppm, it was possible to attribute the two signals at 82.5 and 79.2 ppm to the sugar at N(6), and those at 85.0 and 84.2 ppm to N(2). This difference in the chemical shifts of both pairs of signals [252 Hz for C(1′)–N(6) and 58 Hz for C(1″)–N(2)] is consistent with the fact that, in the latter, the chemical environment is not so different (C=O and SO_2) as in the former (SO_2 and CH_3).

The kinetic parameters were calculated by experimental line-shape analysis using the appropriate equations for the dynamic processes,[12] considering equal populations for each conformer, and taking into account the temperature dependence of the chemical shifts (Table 3). By means of the Eyring equation,[13] the values of activation enthalpy $\Delta H^{\neq}$ and entropy $\Delta S^{\neq}$ were calculated for each process, affording the following average values: for rotation around N(2)–C(1′) $\Delta H^{\neq}$ = 10.2 kcal mol^{-1} * and $\Delta S^{\neq}$ = −16.8 e.u.,† (in good agreement with a previous finding[3]) and for rotation around N(6)–C(1″), $\Delta H^{\neq}$ = 16.7 kcal mol^{-1} and $\Delta S^{\neq}$ = +4.7 e.u. According to this, rotation around the glycosidic bond is less hindered (by 6 kcal mol^{-1}) when the sugar lies between a C=O and an SO_2 group, than between a C=O and a methyl, as expected on the basis of steric criteria.

* 1 cal = 4.184 J. † 1 e.u. = 4.184 J K^{-1}.

Conclusions

Compounds (**5**) and (**6**) provide two new examples of the very few reported cases of restricted *syn*:*anti* rotamers of nucleosides existing in solution, at room temperature. For glucosides of 5-methylthiadiazine, it seems that this is likely to occur when the ring is *N*(2), *N*(6) disubstituted. However, for its pyrimidine analogue, 6-methyluracil, this phenomenon has been observed for some *N*-monosubstituted nucleosides,[14] (3-β-D-glucopyranosyl-6-methyluracil being one of the first reported examples of rotational isomerism[15] although no barriers were calculated). The fact that the SO_2 group lies outside the plane of the molecule, as demonstrated by X-ray data[16] can account for this difference since rotation around the glucosidic bond would be less hindered in thiadiazines than in uracils.

Experimental

The UV spectra were measured with a Perkin-Elmer 402 spectrophotometer. Column chromatography was performed on Merck silica gel 60 (70–230 mesh), and preparative TLC was performed on 20 × 20 cm glass plates coated with a 2 mm layer of silica gel PF_{254} (Merck). The compounds were detected with UV light (254 nm) or by spraying the plate with ethanol–sulphuric acid (3:1) and heating.

^{1}H NMR spectra were obtained on both Varian XL-300 and Bruker AM-200 spectrometers operating at 300 and 200 MHz respectively, using $SiMe_4$ as an internal standard. Typical spectral parameters were: spectral width 10 ppm, pulse width 9 μs (57°), data size 32 K. NOE difference spectra were measured under the same conditions, using a presaturation time of 3 s. 2D-scalar shift-correlated ^{1}H NMR spectra were carried out on Varian XL-300 with the following acquisition parameters: spectral width in both dimensions 1 540 Hz, pulse width 13 μs (90°), relaxation delay 1.5 s, number of increments 128, and 512 × 512 transformed data points.

^{13}C NMR experiments were carried out on the Varian spectrometer operating at 75 MHz. The internal reference was $SiMe_4$ and the temperature was varied in the range 246–328 K. The acquisition parameters were: spectral width 16 kHz, acquisition time 0.99 s; pulse width 9 μs (57°) and data size 32 K.

5-Methyl-6-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-1,2,6-thiadiazin-3(2H)-one 1,1-Dioxide (**3**) *and 2,6-Di-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)-1,2,6-thiadiazin-3(2H)-one*

1,1-*Dioxide* (**6**).—To a solution in dichloromethane (15 cm^3) of the silyl derivative of (**1**)[5] prepared by refluxing the base (1.0 g, 0.006 mol) in hexamethyldisiliazane (20 cm^3) and ammonium sulphate (catalytic amounts) under nitrogen, the glucose derivative (**2**) (2.4 g, 0.006 mol) dissolved in dichloromethane (50 cm^3) was added. The mixture was cooled, and BF_3·Et_2O (3 cm^3) was added with vigorous stirring and exclusion of moisture. The resulting mixture was stirred for 4 h, at room temperature, and was then shaken with saturated sodium hydrogencarbonate solution (100 cm^3). The organic phase was separated, dried over sodium sulphate, and evaporated under reduced pressure. The residue was chromatographed on silica gel column eluting with $CHCl_3$–MeOH (25:1). The first fraction was concentrated and purified by TLC using $CHCl_3$ as the eluant to give (**6**) (0.8 g, 32%) as a colourless glass (Found: C, 46.5; H, 5.3; N, 3.6; S, 3.7. $C_{32}H_{42}N_2O_{11}S$ requires C, 46.71; H, 5.14; N, 3.40; S, 3.90%); λ_{max}(MeOH) 200, 220, and 250 nm (log ε 3.49, 3.62, and 3.85).

The second fraction was purified by TLC eluting with $CHCl_3$–MeOH (10:1) to give (**3**) (1.2 g, 45%) (Found: C, 43.7; H, 4.7; N, 5.75; S, 6.4. $C_{18}H_{24}N_2O_{12}S$ requires C, 43.90; H, 4.91; N, 5.69; S, 6.51%); λ_{max}(MeOH) 202 and 288 nm (log ε 3.49 and 3.83).

6-(β-D-*Glucopyranosyl*)-5-*methyl*-1,2,6-*thiadiazin*-3(2H)-*one* 1,1-*Dioxide* (**4**).—A solution of (**3**) (0.1 g, 0.003 mol) in saturated methanolic ammonia (20 cm^3) was stirred, at room temperature, for 2 h. The solution was evaporated to dryness to yield (**4**) (0.06 g, 92%) of (**4**) as a colourless glass (Found: C, 37.0; H, 5.1; N, 8.2. $C_{16}H_{16}N_2O_8S$ requires C, 37.03; H, 4.97; N, 8.64%); λ_{max}(MeOH) 200 and 290 nm (log ε 3.52 and 3.84).

2,5-*Dimethyl*-6-(2,3,4,6-*tetra*-O-*acetyl*-β-D-*glucopyranosyl*)-1,2,6-*thiadiazin*-3(2H)-*one* 1,1-*Dioxide* (**5**).—A stirred solution of (**3**) (0.25 g, 0.5 mmol) in anhydrous acetone (20 cm^3) was treated with methyl iodide (0.1 cm^3, 0.7 mmol) in the presence of potassium carbonate (0.1 g). The reaction mixture was refluxed for 2 h, and the solid removed by filtration. The solvent was removed *in vacuo* and the residue was chromatographed on preparative TLC using $CHCl_3$–MeOH (25:1) as the eluant to give (**5**) (0.1 g, 43%) as a colourless glass (Found: C, 45.2; H, 5.0; N, 5.6. $C_{19}H_{26}O_{12}N_2S$ requires: C, 45.06; H, 5.17; N, 5.53%); λ_{max}(MeOH) 200, 211, and 265 nm (log ε 3.50, 3.52, and 3.90).

References

1 P. Fernández-Resa, P. Goya, and M. Stud, *Nucleic Acids Res.*, 1978, **4**, 61.
2 P. Goya, A. Martínez, M. L. Jimeno, and W. Pfleiderer, *Liebigs Ann. Chem.*, 1987, 981.
3 C. Ochoa, A. Cuberta, and M. L. Jimeno, *Nucleosides, Nucleotides*, 1987, **6**, 631.
4 W. Saenger, 'Nucleoside Analogues. Chemistry, Biology, and Medical applications,' eds. R. Walker and E. De Clercq, Plenum Press, New York, 1979, p. 1.
5 R. Die, J. Díez, G. García-Muñoz, R. Madroñero, and M. Stud, *J. Heterocycl. Chem.*, 1972, **9**, 973.
6 H. Vorbrüggen and U. Niedballa, *J. Org. Chem.*, 1974, **39**, 3664; *ibid.*, 1974, **39**, 3667.
7 L. B. Townsend, 'Synthetic Procedures in Nucleic Acid Chemistry,' Wiley Interscience, New York, 1973, vol. 2, p. 331.
8 J. Elguero, C. Ochoa, and M. Stud, *Heterocycles*, 1982, **17**, 401.
9 R. Stolarski, L. Dugycz, and D. Shugar, *Eur. J. Biochem.*, 1980, **108**, 111.
10 J. Donohue and K. N. Trueblood, *J. Mol. Biol.*, 1960, **2**, 336.
11 A. Bax, R. Freeman, and G. Morris, *J. Magn. Reson.*, 1981, **42**, 169.
12 F. W. Wehrli and T. Wirthlin, 'Interpretation of C-13 NMR spectra,' Heyden and Son, 1976, p. 197.
13 J. W. Emsley, J. Feeny, and L. H. Sutcliffe, 'High Resolution Nuclear Magnetic Resonance Spectroscopy,' Pergamon Press, Oxford, 1965, ch. 9.
14 E. Wittenburg, *Collect. Czech. Chem. Commun.*, 1971, **36**, 246.
15 K. A. Watanabe, S. S. Saluja, B. A. Otter, and J. J. Fox, *Tetrahedron Lett.*, 1972, **37**, 3911.
16 V. Arán, P. Goya, and C. Ochoa, *Adv. Heterocycl. Chem.*, 1988, **44**, 83.

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