MEASUREMENT OF LONG-RANGE HETERONUCLEAR COUPLINGS : APPLICATION TO OLIGOSACCHARIDE CONFORMATION

Claude Morat,[a] and François R. Taravel[b]

[a] *Laboratoire d'Etudes Dynamiques et Structurales de la Sélectivité, Université Joseph Fourier, B.P. 53 X, 38041 Grenoble cedex, France*
[b] *Centre de Recherches sur les Macromolécules Végétales, CNRS, B.P. 53 X, 38041 Grenoble cedex, France.*

ABSTRACT : $^3J(C,H)$ values have been measured using 2D-J heteronuclear-resolved n.m.r. spectroscopy to estimate interglycosidic conformations in various oligosaccharides with a degree of polymerization ranging from 6 to 30.

One of the most important n.m.r. parameters in conformational studies of all classes of molecules is the vicinal or three-bond coupling constant (^3J) between nuclei like ^{13}C and ^1H in pathways which could involve heteroatoms (oxygen, nitrogen, ...). These couplings show a clear correlation with the dihedral angle θ between the two planes defined by three atoms, as was first noticed by Lemieux et al.[1] and Schwarcz et al.[2] for carbohydrates. This relation is often represented by the general formula :

$$^3J = A \cos^2 \theta - B \cos \theta + C$$

Coefficients A, B and C depend mainly on the nature of the four atoms in the pathway. Such a relation is derived from the first Karplus relation established for H-C-C-H fragments.[3] Its applicability has been proved both theoretically and by measurements on conformationally rigid molecules for a large number of molecular fragments.[4]

In the carbohydrate field, the measurement of such couplings has been obtained through studies of ^{13}C-enriched and selectively deuterated compounds.[5-10] The selective 2D-J heteronuclear experiment proposed by Bax and Freeman[11] and variously used by different authors [12-17] has also been performed on mono-, di- and some oligo-saccharides in natural abundance compounds.

In order to expand the data base (with the ultimate purpose of establishing accurate Karplus-type relationships between $^3J(C,H)$ values and the corresponding dihedral angles) and to obtain experimental data which can be safely used in conformational analysis, we now report our results on a series of oligosaccharides with a degree of polymerization ranging from 6 to 30, using a modified version of Bax and Freeman sequence[15].

The relative orientation of a pair of contiguous sugar residues is described in the

usual way by the torsion angles ϕ and ψ around the glycosidic bonds C1'--O and O--Cn respectively in the fragment H1'--C1'--O--Cn--Hn (primed numbers refer to the glycone and unprimed to the aglycone).

The $^3J(C,H)$ values measured are listed in Table 1. An example of resolution obtained by this technique is shown on Figure 1 with the **1a-1c** compounds. The solvent was CDCl3. The spectra were obtained on Bruker AM 300 or AM 400.
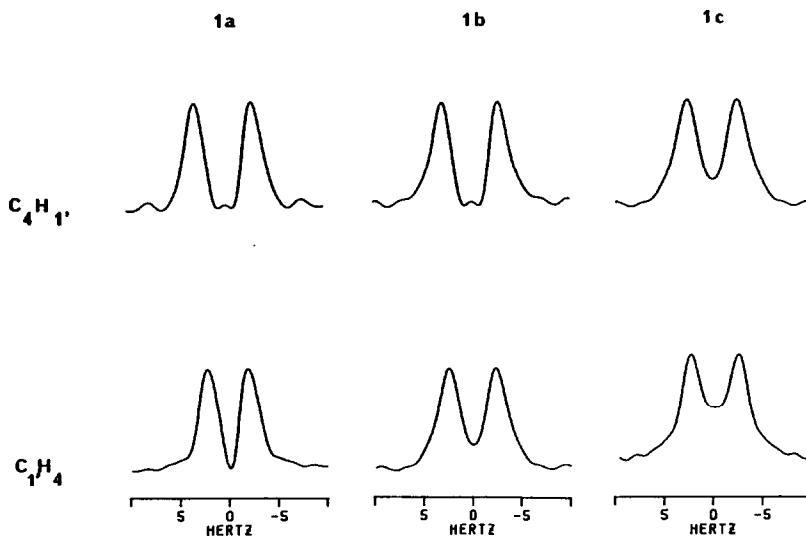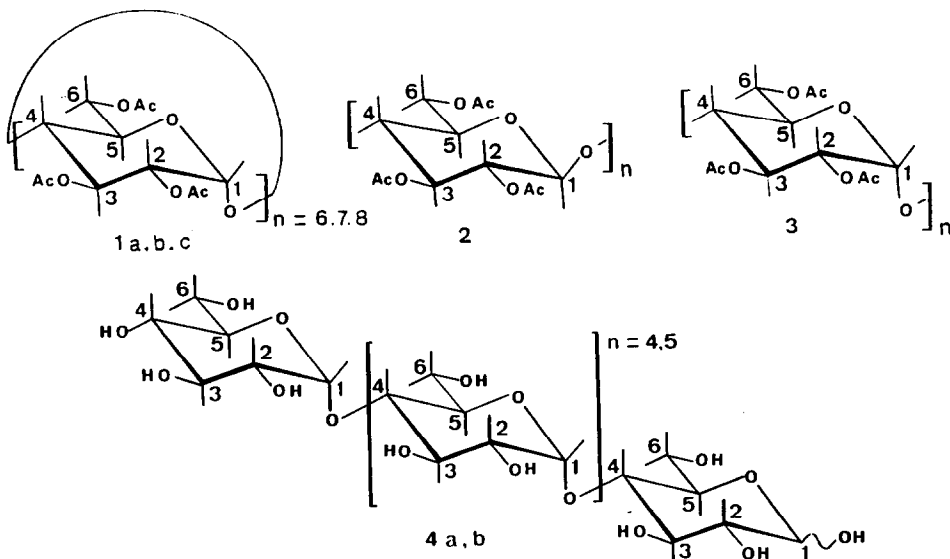

Figure 1 : 2D F1-dimension slices of C-1 and C-4 corresponding to H-4 (bottom) and H-1' (top) selective inversion on compounds **1a, 1b** and **1c**. Coupling constant values have been measured without any correction.

These values were used to estimate the torsion angle about the glycosidic bond via an equation with the following parameters A = 5.7, B = 0.6, C = 0.5. These were established for a series of conformationally rigid carbohydrate derivatives[18] and they lead to a Karplus-type curve which fits fairly well our values obtained previously[15]. They are also very close to the parameters used by B. Mulloy *et al.*[17].

The torsion angles determined for the acetylated cyclomaltodextrines are all in the region 0-35° as shown below.

Interglycosidic carbon-proton coupling constants for oligosaccharides.

Compound	Linkage	$^3J_{C,H}$	Value(Hz)[a]	Estimated Torsion angle[b]
Peracetate cyclomaltohexaose (**1a**)	α(1,4)	C-4,H-1'	5.6	0
		C-1,H-4	4.0	33
Peracetate cyclomaltoheptaose (**1b**)	α(1,4)	C-4,H-1'	5.5	0
		C-1',H-4	4.6	25
Peracetate cyclomaltooctaose (**1c**)	α(1,4)	C-4,H-1'	4.8	22
		C-1',H-4	4.5	26
Cellulose triacetate (DP 30) (**2**)	β(1,4)	C-4,H-1'	5.6	0
		C-1',H-4	5.7	0
Amylose triacetate (DP 30) (**3**)	α(1,4)	C-4,H-1'	2.9	45
		C-1',H-4	4.6	25
Maltohexaose (**4a**)	α(1,4)	C-4,H-1'[c]	4.3	29
		C-1',H-4[c]	5.0	19
Maltoheptaose (**4b**)	α(1,4)	C-4,H-1'[c]	4.4	28
		C-1',H-4[c]	5.1	18

[a] Coupling constants are given at ± 0.5 Hz. This lead to estimate the torsion angle to ± 10°.

[b] As several solutions exist for a given coupling constant only the absolute value in the zero region is presented.

[c] Coupling constants corresponding to central residues

They show a slight variation of the torsion angles in parallel to the increase of the annular aperture in going from the cyclic hexasaccharide (**1a**) to the octasaccharide (**1c**). This is characterized by a great similarity between the cyclic hexa- and heptasaccharides, as already mentioned by Lindner *et al.*[19] for the OH-form of the cyclomaltodextrines, and also by an adjustment of the φ torsion angle for the octasaccharide in order to assume a larger radius of the macrocycle.

The conformation of cellulose triacetate (**2**) in CDCl$_3$ can be described by torsion angles close to zero. This result is consistent with the torsion angles found in the solid state (φ = 15°, ψ = -22°) from X-ray and electron diffraction analyses.[20] These lead to the presence in solution of chains with a two-fold axis.

The results for amylose triacetate (**3**) could indicate that in solution the molecule has a relatively high degree of freedom as the dihedral angles estimated in our study differ from

those published for the crystalline structure[21] (-39° and -50° for the ϕ and ψ angles respectively).

At last within the accuracy of our data, the coupling constants corresponding to the central residues for maltohexaose (**4a**) and maltoheptaose (**4b**) are similar indicating a very close conformation for these two oligosaccharides of the amylose family.

References

1.   R.U. Lemieux, T.L. Nagabhushan, and B. Paul, *Can. J. Chem.*, 1972, **50**, 773.

2.   J.A. Schwarcz and A.S. Perlin, *Can. J. Chem.*, 1972, **50**, 3667.

3.   M. Karplus, *J. Chem. Phys.*, 1956, **30**, 11.

4.   P.E. Hansen, *Progress in NMR Spectroscopy*, 1981, **14**, 175.

5.   A. Parfondry, N. Cyr, and A.S. Perlin, *Carbohydr. Res.*, 1977, **59**, 299.

6.   G.K. Hamer, F. Balza, N. Cyr, and A.S. Perlin, *Can. J. Chem.*, 1978, **56**, 3109.

7.   K. Bock and C. Pedersen, *Acta Chem. Scand., Ser. B*, 1977, **31**, 354.

8.   T.E. Walker, R.E. London, T.W. Whaley, R. Barker, and N.A. Matwiyoff, *J. Am. Chem. Soc.*, 1976, **98**, 5807.

9.   D.Y. Gagnaire, R. Nardin, F.R. Taravel, and M.R. Vignon, *Nouv. J. Chimie*, 1977, **1**, 423.

10.   S. Perez, F.R. Taravel, and C. Vergelati, *Nouv. J. Chimie*, 1985, **9**, 561.

11.   A. Bax and R. Freeman, *J. Am. Chem. Soc.*, 1982, **104**, 1099.

12.   M.J. Gidley and S.M. Bociek, *J. Chem. Soc., Chem. Commun.*, 1985, 220.

13.   G. Batta and A. Liptak, *J. Chem. Soc., Chem. Commun.*, 1985, 368.

14.   F.H. Cano, C. Foces-Foces, J. Jimenez-Barbero, A. Alemany, M. Bernabé, and M. Martin-Lomas, *J. Org. Chem.*, 1987, **52**, 3367.

15.   C. Morat, F.R. Taravel, and M.R. Vignon, *Magn. Reson. Chem.*, 1988, **26**, 264.

16.   C. Morat and F.R. Taravel, *Tetrahedron Lett.*, 1988, **29**, 199.

17.   B. Mulloy, T.A. Frenkiel, and D.B. Davies, *Carbohydr. Res.*, 1988, **184**, 39.

18.   I. Tvaroska, M. Hricovini, and E. Petrakova, *Carbohydr. Res.*, 1989, **189**, 359.

19.   K. Lindner and W. Saenger, *Carbohydr. Res.*, 1982, **99**, 103.

20.   E. Roche, H. Chanzy, M. Boudeulle, R.H. Marchessault, and P. Sundararajan, *Macromolecules*, 1978, **11**, 86.

21.   A. Sarko and R.H. Marchessault, *J. Am. Chem. Soc.*, 1967, **89**, 6454.